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THE FECAL BACTERIA OF HEALTHY MEN.*

PART I. INTRODUCTION AND DIRECT QUANTITATIVE OBSERVATIONS.†

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QUANTITATIVE RESULTS.—The number of bacteria in feces. The weight of bacterial dry substance in feces. The relation of these to each other. The daily excretion of bacteria. Fecal nitrogen contained in the bacteria. The variety of fecal bacteria recognizable by microscopic examination.

SUMMARY.

REFERENCES.

INTRODUCTION.

THE fundamental importance of bacteria in the physiology of the digestive tract of vertebrates is quite generally recognized. The researches of Nuttall and Thierfelder,[18] Schottelius,[21] and Mme.

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† The culture experiments will be reported in Part II, which will appear in a short time.

Metchinkoff[17] still leave unsettled the question, suggested by Pasteur,[19] whether animal life could continue without their presence. For the pathologist, intestinal bacteriology has long been a fertile field and the researches relating to it are very numerous indeed. We shall not attempt a citation or any review of the voluminous literature of this subject as these may be found in the monograph on the micro-organisms of the feces by Strasburger.[20] Only the researches of others which are closely related to our own work will be considered briefly in the description of our methods and results.

During the past year this laboratory undertook an experiment to determine the influence of cured meats upon human health. In connection with this experiment it was deemed advisable to study the fecal flora, inasmuch as this study might prove of value as an index to the condition of the digestive tract and of possible alterations due to different diets. It was also recognized that here was afforded a somewhat exceptional opportunity for the study of the micro-organisms of the feces of normal adult men during a long period of time. No very comprehensive examination of the fecal flora in a group of normal adults seems to have been previously undertaken.

It is our purpose to present, in this communication, the methods of examination employed, together with the results obtained, wherever these appear to be of value as a contribution to the physiological or pathological bacteriology of the intestine. During the course of the work 266 stools were examined. About three-fourths of these serve as the basis of this paper.

Source of Material.

The observations were made upon 12 men, university students from 19 to 30 years of age, living together in a club-house specially provided for the metabolism experiment. They were fed exclusively at the club table upon weighed quantities of foods, the composition of which was determined by chemical analysis. The menu provided was similar to that of a student boarding-house or club of the better grade, with considerable variety of diet from day to day. The breakfast consisted of fruit, cereal, meat, potatoes, bread, butter, water, and milk or coffee. For luncheon, a soup, cereal, fruit, bread, butter, water, and milk or tea were served. Dinner was served in the evening. It consisted of meat, potatoes, a second vegetable, a pudding, bread, butter, water, and milk or cocoa. For the sake of variety, various fruits, cereals, soups, vegetables, and meats were used, and the potatoes were prepared in various ways for the different meals. These variations were all made in accordance with a definite schedule of meals. This same schedule was exactly repeated every eight days. A certain amount of selection was allowed to each individual subject, as to the quantity of the different foods he should consume, as long as this choice did not interfere with the proper relation between food intake and excretion. For example, Subjects K and L were the only ones who chose to take coffee; Subjects H and I did not eat the second vegetable dish at dinner after March 16; Subject E ate only one-half the usual amount of meats after January 12, and Subject F, only three-fourths the usual amount of meats during the whole period of observation; Subject B ate none of the cereals. These variations are such as would ordinarily occur on account of individual tastes. There was also a variation in the quality of the meats consumed by the different subjects during a part of the period of observation. Some of the subjects during this time received cured meats as part of the meat ration, while others received only perfectly fresh meats during the entire period of observation. The quantity of cured meats and of meat preservative consumed was in no case greater than that which may ordinarily be consumed in boarding-houses or clubs. At least half of the meats served to each individual were fresh and free from preservatives. Thus the diet with its variations represents fairly well an ordinary mixed diet.

A detailed discussion of the diet would lead beyond the scope of the present paper. It is not our purpose to consider the relation of the fecal flora to variations in the diet, this question being reserved for another time. For the present, the variations in diet may be regarded as normal and the results of the bacteriological examinations regarded as fairly representative of normal intestinal conditions.

PLAN OF THE WORK.

Our observations fall in general under two heads: first, the direct examination, including the quantitative determination of the bacteria in the feces by microscopic enumeration and by the gravimetric method of Strasburger,[23] and the determination of the relative numbers of morphologically different bacteria present by microscopic study of Gram stained films; second, the culture experiments, which were carried out in a quantitative manner in each case, (*a*) to determine the number of bacteria capable of development on various media in the air and in a hydrogen atmosphere, and to identify these species as far as possible; (*b*) to determine the fermentative activity of the mixed fecal flora upon various sugars, as suggested by Herter;[8] (*c*) to bring to development by special methods the different species of bacteria present, which fail to develop upon ordinary plates.

COLLECTION OF MATERIAL.

Each stool was passed directly into a sterilized agateware basin, provided with a cover, no special precautions being taken to avoid contamination during passage through the anal canal, but all contamination of the material after passage being carefully excluded. As a rule the material was delivered at the laboratory within ten minutes after passage and the examination of it begun at once. In exceptional cases immediate examination was impossible and then the material was kept cold by standing the pan on a block of ice in the ice box until examined.

PREPARATION.

The weighing, macroscopic examination, and thorough mixing of the feces were performed as rapidly as possible. Ordinarily a fairly good mixture was obtained by rubbing the feces with a sterile porcelain spatula in the collecting basin. At times the material was transferred to a large mortar and mixed with a pestle. The difficulty experienced in getting a satisfactory mixture varied considerably with different stools, and in no case could the final mixture be regarded as very uniform, as macroscopic bits of vegetable tissue 1 to 5 mm. in length commonly remained undivided. However, the error due to this factor was not very serious because the sample taken for examination was fairly large.

Bacteriological Suspension.

Immediately after the mixing was completed a 1:100 suspension of the feces was prepared to serve as the basis for the general bacteriological examination. This required considerable care in its preparation but the method we have used is very simple and the resulting suspension uniformly satisfactory. In order to get a uniform suspension of the fecal bacteria it is important that sufficient diluent be used. The suspension of 5 gm. of feces in 50 c.c. of liquid we found to be very difficult but the suspension of one-half gm. feces in 50 c.c. was found to be a relatively simple matter. The procedure is as follows: A perfectly clean, dry, sterilized, glass-stoppered measuring flask of 50 c.c. capacity is first accurately weighed on the analytical balance. Then by means of a sterile glass rod about 4 mm. in diameter and somewhat pointed at the tip, 500 mg. mixed feces is transferred to the flask and deposited on the inside of the neck, the quantity being subsequently increased or diminished as necessary until the required amount is obtained. The weight is observed with the glass stopper in position and an amount of feces within 1 mg. of the desired weight is considered sufficiently accurate. Rapid manipulation is essential to avoid error from loss by evaporation. About 10 c.c. of sterile 0.75 per cent salt solution is now transferred to the flask by means of a Pasteur bulb pipette and the feces rubbed up in the neck of the flask with another clean sterile glass rod. Further salt solution is then added, the glass rod and the neck of the flask being washed with it as it runs in, until the flask contains about 20 c.c. The flask is then stoppered and vigorously shaken for a minute, then inverted so that unsuspended solid particles settle into the neck against the glass stopper. The flask is then slowly erected so that the liquid is decanted from the neck back into the body of the flask, leaving the solid bits in the neck where they are now easily crushed with the glass rod. The flask is now again shaken and the solid particles again allowed to settle into the neck, this procedure being repeated until there remain no macroscopic bacterial masses, recognized as opaque brown lumps. The visible pieces of tissue will then be washed clean so that their character is easily recognized and the bacteria have been washed away and suspended in the liquid. The flask is now violently shaken for 3–5 minutes to break up microscopic bacterial clumps. Further salt solution is then added up to the 50 c.c. mark, the glass rod and the neck of the flask being washed clean during the process, and after a final vigorous shaking the suspension is ready for use. It must always be thoroughly remixed immediately before measuring out portions from it.

The preparation of such a suspension requires 10 to 20 minutes of careful work but after a little practice the result is uniformly satisfactory, and microscopic examination shows the individual bacteria well separated. This suspension is used for the enumeration of the total bacteria, for the differential counting of the different types, and for all the culture experiments. One c.c. of it represents the bacterial content of 10 mg. feces.

QUANTITATIVE DETERMINATIONS

As is well known the plate method is of no value for the estimation of the total quantity of bacteria present in feces, as the number of bacteria here capable of development on plates is only a small fraction of the total quantity present, and bears no constant relation to the latter number. The total quantity of bacteria was determined by three separate methods, the microscopic counting method of Winterberg,[25] the Eberle[5]-Klein[10] counting method, and the gravimetric method of Strasburger.[23]

THE EBERLE-KLEIN METHOD.

Eberle,[5] working in Escherich's laboratory, was the first to determine the quantity of fecal bacteria by counting them in stained films. His experiments were carried out with normal infants' stools. Coverglass preparations were made from fecal suspensions of known dilution and after being completely dried were stained with freshly prepared aniline water fuchsin, then washed in water, air-dried, mounted in Canada balsam, and counted.* The method was very much improved by Alex. Klein[10] and Hehewerth.[6] Klein had noticed that vegetative bacteria were more sensitive to disinfectants in a moist, than in a dry state, and this led him to believe that bacteria would also be stained more readily in a moist condition than after drying. Therefore he allowed the dye to act while the bacteria were suspended in a liquid and the coverglass preparation was made, dried, and fixed, only after the bacteria were stained. Later the same investigator[12] modified the method still further, by using gelatin to fix the stained bacteria to the coverglass. The aqueous solution of gelatin and the stained bacterial emulsion were put upon the coverglass separately, then mixed and spread. When dry the preparation was immediately mounted in Canada balsam without flaming.

The procedure as we have employed it has been considerably modified. In the addition of the gelatin to the bacterial suspension, in spreading the films, in selecting the fields to be counted, and in counting individuals, rather than groups, as units, the technic differs from Klein's method. Of the 1:100 suspension of the feces prepared as described above (p. 126), 2½ c.c. is transferred to a clean dry bottle, ½ c.c. of melted nutrient gelatin and 2 c.c. of aniline water gentian violet added, and the whole thoroughly mixed and allowed to stand for 3–5 minutes. Then by means of a platinum loop, the carrying capacity of which has been previously determined with great care, a loopful of the mixture, well shaken immediately before, is transferred to a clean, flamed 20 mm. square No. 1 coverglass and deposited near the center of the glass. Immediately another coverglass of the same size is accurately placed on top of the first so that the two glasses

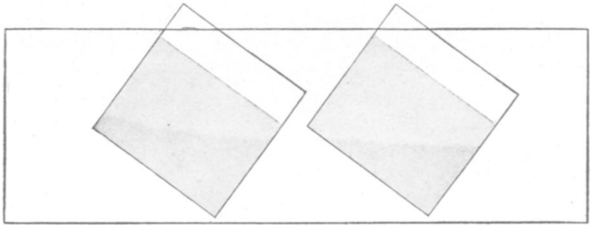

FIG. 1.—Coverglasses mounted so that the proper diagonals of the bacterial films lie in the longitudinal diameter of the slide.

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* Winslow[24] has employed a similar method in estimating the number of bacteria in sewage.

are in contact over about ¾ of their surfaces and the sides evenly fitted together. As soon as the liquid has spread evenly between the two coverglasses, they are quickly slipped apart and allowed to dry. The technic here is the same as ordinarily used in the preparation of coverglass blood films. If the preparations do not appear evenly spread the process is to be repeated with two more coverglasses until a satisfactory result is obtained. The films are next accurately measured by a millimeter rule and then, without further treatment, mounted in Canada balsam upon one slide so that one diagonal of each rectangular film is parallel with the long axis of the slide. By laying the slide over co-ordinate paper these diagonals are readily brought into the same line parallel with the edge of the slide, as shown in Fig. 1.

The covers must be so mounted that these diagonals now in a straight line are those which crossed each other as the two coverglasses were originally placed together in preparing the films. When properly made each diagonal measures almost exactly 25 mm.

For counting the bacteria in the preparation, the Leitz $\frac{1}{12}$ oil immersion objective and the No. 3 ocular fitted with an Ehrlich ocular square to restrict the field to a convenient size, and a mechanical stage graduated in millimeters are employed. Beginning at the end of the diagonal of one coverglass the bacteria are counted in each of 25 fields 1 mm. apart along this diagonal. In a similar way 25 fields are counted on the diagonal of the second coverglass, making a total of 50 fields, the average of which may be considered as representative of both films. The size of the square field is accurately measured by a stage micrometer. From the data then at hand the number of bacteria per milligram feces is calculated.

Example.—B 252, Subject H, July 15, 1908. In the preparation made as described each film measures 16.5×17.5 mm., total film area therefore, 33×17.5 mm.; the field employed measured 0.0445 mm. square; the amount carried by the loop 2.01 mg.; the number of bacteria counted in 50 fields was 559 and the original 1:100 suspension was used (diluted to 1:200 by dye and gelatin). From these data,

$$\text{Bact. per mg. feces} = \frac{559 \times 33 \times 17.5 \times 200}{50 \times 0.0445 \times 0.0445 \times 2.01} = 324{,}000{,}000.$$

In this fraction all the members except the size of the films and the number of bacteria counted may be kept constant and the calculation simplified by use of logarithmic tables.

With apparatus and reagents ready and the 1:100 suspension prepared, this entire estimation can be completed in about 40 minutes. The results cannot be considered very accurate as the platinum loop does not carry an exactly constant quantity. There is also sometimes great difficulty in distinguishing micrococci from other fine particles in the preparations. The concentration of the bacterial suspension also influences the final result. The estimation is in general relatively higher when dilute suspensions are counted as we have observed in applying it to enumeration of bacteria in pure cultures.

THE WINTERBERG METHOD.

The first employment of the Thoma-Zeiss blood-counting chamber for bacteriological technic was made in the one-cell dilution method of obtaining a pure culture. By this method the number of cells per unit volume of a suspension could be ascertained and the dilution required to obtain a suspension with one cell in two to five drops calculated.[9] In this way the first pure bacterial culture was obtained.[13] Heinrich Winterberg[25] was the first to use this method of bacterial counting, and to test its accuracy both as regards suspensions of varying dilutions and in comparison with the microscopic-plate counting method. He considers the method more accurate than the plate counting method, but as a quantitative procedure he would consider it unimportant. He says his determinations were too low. Winterberg's counts were made with suspensions of living bacteria in bacteria-free distilled water.

The method, as we have used it, is as follows: a portion of the 1:100 suspension of feces is diluted 10 times and a portion of this is drawn up to the mark 1 in the capillary of a diluting pipette, ordinarily used in estimating the white blood cells. This is diluted to the mark 11 with a dilute solution of methylene blue in physiological-salt solution. (The staining solution consists of methylene blue 1 gram, glycerin 25 c.c., distilled water 75 c.c. A few drops of this are mixed with 10 c.c. of 0.8 per cent salt solution until the mixture is well colored but not too opaque. This mixture is used as the diluting fluid. A little practice will show the proper depth of color to be employed.) The suspension is thoroughly mixed in the bulb by shaking and rolling in the usual manner. Several drops are blown out and then a very small drop is placed in the center of the circular elevated portion of the slide, which has been previously thoroughly cleaned by washing in distilled water and alcohol. A clean, thin, ground cover-slip is made slightly moist by breathing upon it and is immediately placed upon the slide. Slight pressure upon it causes the Newton color rings to appear and these remain after the pressure is removed if the preparation has been properly made. The slide is allowed to stand one to two hours to allow the bacteria to settle. Then the bacteria in 50 small squares upon the marked scale are counted microscopically, the No. 7 Leitz objective and No. 3 ocular being used. The calculation is simple as an example will show.

Example.—B 252, Subject H, July 15, 1908. 50 squares contain 400 bacteria. Therefore the average per square is 8.0 bacteria. One small square $=\frac{1}{4000}$ c.mm. One c.mm. contains $8\times4000=32{,}000$ bacteria. One c.c. contains $32{,}000\times1{,}000=32{,}000{,}000$ bacteria. Pipette dilution = 1:10. $32{,}000{,}000\times10=320{,}000{,}000$ bacteria per c.c. of $\frac{1}{1000}$ suspension. One c.c. of $\frac{1}{1000}$ suspension is equivalent to 1 mg. feces. Therefore there are 320,000,000 bacteria per mg. feces.

Counting by this method is an exacting process and much practice is required to see all the bacteria present. Careful adjustment of the light is important and best results have been obtained by illumination with the Welsbach light; constant focusing through the different layers is necessary. Perhaps the greatest source of error is the difficulty of distinguishing accurately the bacteria. Skill in this is acquired only by considerable practice. The method has the advantage of simplicity. Our results seem to confirm those of Winterberg in that high dilutions give a relatively high count.

GRAVIMETRIC DETERMINATION.

The quantitative determination of the bacteria by the gravimetric method is essentially the procedure of Strasburger.[23] Immediately after making up the 1:100 suspension this determination is begun. To determine the dry substance we weigh out quickly two portions (2 to 3 gm.) of feces in weighed porcelain crucibles and dry to constant weight at 102° to 103° C. Two similar amounts (2 to 3 gm.) are also weighed out in weighed centrifuge tubes. The weighings are done as rapidly as possible and read accurately within two milligrams. Our method here differs from that of Strasburger, who employs a burette to measure off exactly 2 c.c. for each determination, and from that of Steele[22], who uses a pipette to measure off 5 c.c. of a homogeneous suspension of the whole stool diluted with salt solution. We also consider it quite desirable to do the determinations, both of the dry substance and of the bacteria, in duplicate. Weighing the material in the centrifuge tubes saves time and avoids loss in transferring. Each tube is carried through as a separate determination.

The bacteria are separated from the remaining fecal substance by a process of fractional sedimentation in the centrifuge. To the feces in the centrifuge tube a few drops of ½ per cent hydrochloric acid are added and mixed to a smooth paste by means of a glass rod. Further amounts of the acid are added with continued stirring until the material is thoroughly suspended and the tube nearly full. The tube is then whirled in the centrifuge at high speed for ½ to 1 minute. We have found the electric centrifuge to be the most satisfactory. By this treatment the suspension is sedimented into more or less definite layers, the uppermost of which is fairly free from the larger particles while it still contains a considerable quantity of the bacteria. The upper three-fourths of the suspension are now drawn off by means of a Pasteur bulb pipette and transferred to a beaker properly labeled. The sediment remaining in the tube is again rubbed up with the glass rod, which is kept in the beaker during the centrifugation, with the addition of further dilute acid, and again centrifugated for ½ to 1 minute. The supernatant liquid is pipetted off and added to the first, the same pipette being used for the one determination throughout. A third portion of the dilute acid is then added to the sediment, again mixed by stirring and by vigorous shaking, and again centrifugated. In this way a third, fourth, and eventually a fifth washing is obtained, all being added to the first. During the process care is taken to wash the material from the walls and mouth of the centrifuge tube down into it. Finally when the sediment is sufficiently free from bacteria, the various remaining particles are visibly clean,* and the supernatant liquid after centrifugation remains almost clear. The pipette is thoroughly rinsed out with this last portion which is then added to the preceding fractions in the beaker. We now have in the beaker practically all the bacteria present in the original portion of feces and in the centrifuge tube a considerable amount of the other solid matter. The latter is discarded. The suspension in the beaker is now transferred to clean centrifuge tubes and centrifugated at high speed for a minute and the supernatant liquid transferred to a clean beaker by means of the pipette. The tubes are then refilled from the first beaker and thus all the suspension centrifugated a second time. The first beaker is finally carefully washed with the aid of a rubber-tipped glass rod, the second sediment in the centrifuge tubes washed free of bacteria by means of this wash water and by successive portions of dilute acid, and the supernatant liquid after centrifugation added to the contents of the second beaker. The second clean sediment is discarded. The bacterial suspension now in the second beaker is again centrifugated in the same way and a third portion of bacteria-free sediment separated and discarded. Frequently a fourth serial centrifugation is performed, always if the third sediment is of appreciable quantity. It will be noted that the method of separation is one of fractional sedimentation, differing somewhat from the procedure as practiced by Strasburger who centrifugates only twice in series, and discards the second sediment without washing it, and differing also from that of Steele who centrifugates once and then filters the suspension through muslin to remove the non-bacterial solid matter still remaining in it. We believe that our method gives more accurate results and that the final suspension contains less non-bacterial matter.

At all stages of the separation small portions of the dilute hydrochloric acid should be used for the washings, so that the final suspension shall not be too voluminous. Ordinarily it amounts to 150 to 300 c.c.

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* In case of uncertainty, microscopic examination should be resorted to. This is especially necessary before one has become familiar with the method.

To the final bacterial suspension an equal volume of alcohol is added and the beaker set aside to concentrate. For this purpose Strasburger set the dish into a water bath at 40° C. We employ a bacteriological incubator regulated at a temperature of 50° C. in which the evaporation of the liquid is accomplished in about a week. When concentrated to about 50 c.c. the beaker is removed, about 200 c.c. of alcohol added, covered, and allowed to stand in the room 24 hours. At the end of this time the bacterial substance is generally settled so that most of the clear supernatant liquid can be directly siphoned off without loss of solid matter. The remainder is then transferred to two centrifuge tubes, centrifugated, and the remaining clear liquid pipetted off and discarded. The sediment is next treated with absolute alcohol, mixed with the alcohol and allowed to stand from ½ to 1 hour; then centrifugated and the alcohol removed. The tubes are now filled with ether, the sediment stirred up with a glass rod, the tubes corked and set aside for 24 hours. The cork is held in position by passing a rubber band over it and several times around the lower tapering end of the tube.

On the following day the ether is removed, the sediment washed with absolute alcohol, with aid of the centrifuge and by stirring with a glass rod, and then transferred to a weighed porcelain crucible by means of 95 per cent alcohol. The tube is carefully cleaned with a rubber-tipped rod and alcohol, so that all the sediment is finally in the crucible. With a little care and practice one can get all the sediment transferred to a small No. 00 crucible. Exceptionally it may happen that the crucible becomes full before the tube is clean, in which case the former is placed in the oven and allowed to dry down somewhat before adding the last washing. The crucible is next placed at 50° C. until the excess of liquid has evaporated and then dried at 102° for 48 hours, cooled in a dessicator, and weighed. It is then replaced at 102° C. and weighed again on each succeeding day until the weight is constant within 1 mg. From the data thus determined the percentage of dry bacterial substance in the moist feces and the percentage of bacterial substance in the dry feces are calculated. By applying the factors obtained by a series of such determinations to the total amounts of feces excreted over a considerable period, the average weight of dry bacterial substance excreted daily has been calculated.

NITROGEN.

The nitrogen in the bacterial residues was determined by the Kjeldahl method. This served as a most valuable check upon the purity of the bacterial residue. In general we rejected as unsatisfactory those Strasburger determinations the residues of which contained less than 10 per cent of nitrogen. Some residues contained as much as 12½ per cent of nitrogen. Undoubtedly there is considerable variation in the nitrogen content of different bacterial cells and of cells of the same species under different conditions of growth as has been clearly shown by Cramer.[2] We have found, however, that when the high-speed electric centrifuge was employed the nitrogen content fell below 10 per cent so rarely that technical error was always suspected when it did occur.

From the nitrogen factors thus obtained we have calculated the average daily bacterial nitrogen in the feces and the percentage of total fecal nitrogen represented by it. This ratio between bacterial nitrogen and total nitrogen of the feces would seem to be a fairly constant one under normal conditions when a uniform diet is being taken, and its determination in pathological conditions may prove of value. It can, of course, be determined from a single stool and does not require the separate collection and weighing of the feces for definite periods, which is necessary for the estimation of the quantity of daily bacteria by the Strasburger method.

Example.—Strasburger determination—No. 252, Subject H, July 15, 1908.

(*a*) Dry substance.

	Fresh	Dry
(1) Crucible No. 8 + feces	10.3000 gm.	7.5234 gm.
Crucible No. 8	6.9561	6.9561
Feces	3.3439	0.5673
(2) Crucible No. 12 + feces	10.3965 gm.	7.6449 gm.
Crucible No. 12	7.0913	7.0913
Feces	3.3052	0.5536

Dry substance per gram feces = (1) $\frac{0.5673}{3.3439} = 0.16965$ gm.

(2) $\frac{0.5536}{3.3052} = 0.16749$

Average (1 and 2) = 0.16857

(*b*) Dry bacteria.

(1) Centrifuge tube + feces	10.0206 gm.
Centrifuge tube	7.7316
Feces taken for separation of bacteria	2.2890
Crucible No. 14 + separated bacteria, dry	7.9516 gm.
Crucible No. 14	7.8362
Dry bacteria	0.1154
(2) Centrifuge tube + feces	9.2812 gm.
Centrifuge tube	7.4363
Feces taken for separation of bacteria	1.8449
Crucible No. 15 + separated bacteria, dry	7.0990 gm.
Crucible No. 15	7.0063
Dry bacteria	0.0927

Dry bacteria per gram feces = (1) $\frac{0.1154}{2.2890} = 0.05042$ gm.

(2) $\frac{0.0927}{1.8449} = 0.05025$

Average (1 and 2) = 0.05033 gm.

(*c*) Dry bacterial substance in dry feces $= \frac{0.05033}{0.16857} = 29.86$ per cent.

(*d*) Nitrogen in bacterial residue.

(1) Crucible No. 14 − 0.013419 gm. of nitrogen.
(2) Crucible No. 15 − 0.010823 " " "

Nitrogen in dry bacteria = (1) $\frac{0.013419}{0.1154} = 11.63$ per cent.

(2) $\frac{0.010823}{0.0927} = 11.67$ per cent.

Average (1 and 2) = 11.65 per cent.

(*e*) Average dry bacterial substance excreted daily.
Average daily dry substance of feces of subject during this eight-day period = 20.508 gm.
Average daily dry bacterial substance = 20.508 × 29.86 = 6.13 gm.

(*f*) Ratio of bacterial nitrogen to total fecal nitrogen.
Average daily nitrogen of feces of subject at this period = 1.495 gm.
Average daily bacterial nitrogen = 11.65 per cent of 6.13 = 0.717 gm.

$$\text{Bacterial in total nitrogen} = \frac{0.717}{1.495} = 48.0 \text{ per cent.}$$

DIFFERENTIAL COUNT.

In estimating the relative numbers of morphologically different bacteria of the feces, thin coverglass film preparations of the 1:100 suspension were employed. These were stained by a modified Gram's method,[20] thus adding to the different forms the further distinction afforded by this stain. The technic of the method is as follows:

1. Make a rather thin, even coverglass film preparation.
2. Dry in the air and fix in the flame.
3. Float the coverglass on anilin water gentian violet three minutes.
4. Drain off the excess of dye with filter paper, without washing.
5. Float on Lugol's solution two minutes.
6. Drain and dry the preparation between filter papers.
7. Decolorize in anilin-xylol by immersion for one-half minute.
8. Dry between filter papers.
9. Decolorize further in absolute methylic alcohol for about five seconds.
10. Wash in water.
11. Counterstain with a dilute aqueous solution of fuchsin for a few seconds.
12. Wash in water, blot, dry in the air, and mount in rectified balsam.

The anilin water gentian violet is freshly prepared by adding 0.5 c.c. saturated alcoholic solution of gentian violet to 10 c.c. of saturated aqueous solution of anilin. The Lugol's solution used contains 1 gm. iodine and 2 gm. potassium iodide in 300 c.c. distilled water. The anilin-xylol is made by mixing anilin, one part, with xylol, four parts. In the stained preparation five hundred bacteria are counted and classified according to form and color.

Considerable difficulty was experienced in accurately differentiating Gram negative and Gram positive forms and it cannot be doubted that bacteria of the same species were red in some preparations and blue in others, while it is equally certain that different parts of the same coverglass frequently showed these variations. Sometimes it was very difficult or even impossible to distinguish clearly between the bacteria and minute bits of other material present in the preparation. When we remember the large proportion of fecal bacteria already dead and disintegrating the difficulty of differential staining is not surprising. The procedure has however seemed to us to be of some value in the study of feces of adults.* Its value in the case of infants is already well known.

QUANTITY OF FECAL BACTERIA.

It has long been known that a considerable portion of the substance of the human feces consists of bacterial cells. Eberle[5], working in Escherich's laboratory, found an average of 33,000,000 bacteria per milligram feces, in a two-months-old, milk-fed infant. Hellström[7] studied the fecal flora of the new-born, both by the microscopic and by the plate method. He found that the number of bacteria in the feces increased very rapidly after birth and by the end of the fourth day had become 40,000,000 bacterial cells per milligram feces, a result very close to that of Eberle. Cornelia de Lange[4] studied the fecal flora of infants, employing the counting method of Klein. In a series of cases, she found values from 42,000,000 to 932,000,000 cells per milligram dry substance, or from 11,000,000 to 298,000,000 bacteria per milligram of fresh feces, and concluded that the number of fecal bacteria in milk-fed infants is less than 1,000,000,000 per milligram fecal dry substance.

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* Cohendy, *Comptes rendus Soc. de Biol.*, 1906, 60, p. 415, gives a short notice of a similar study.

Klein[11] counted the bacteria in the feces of adults and in a series of fourteen examinations found from 20,162,000 to 165,614,000 bacteria per milligram, and an average of 58,800,000 per milligram fresh feces. The number of fecal bacteria excreted in 24 hours, he found to be, minimum, 30×10^{11}, maximum, 248×10^{11}, average 88×10^{11}. He estimated* the average weight of the daily bacterial excretion as 293 mg., the bacterial portion of the dry substance as 0.13 per cent, and the bacterial nitrogen excreted daily in the feces as 4.39 mg. Strasburger[23] found by his gravimetric method that about one-third of the dry substance of normal adult human feces consists of bacterial substance. In a series of 16 cases, most of them pathological, he obtained values from 17.2 per cent to 68.4 per cent. The daily quantity of dry bacterial substance in this series varied from 2.6 gm. to 20.0 gm. Three of the subjects were normal individuals, and in these cases the daily bacterial dry substance (average of three days) was 11.8, 6.6, and 5.8 gm., giving an average of 8.1 gm. From this quantity he calculated the number of bacteria excreted daily to be 128×10^{12}, a number about 15 times as great as that of Klein. He did not enumerate the bacteria by the microscopic method. Strasburger mixed the bacterial residues from his several determinations and estimated the nitrogen content of the mixture in duplicate, obtaining 10.364 and 10.305 per cent of nitrogen. From this he concluded that more than half the fecal nitrogen is contained in the bacteria. Lissauer[15], using Strasburger's procedure, determined the quantity of bacteria in 19 different stools from eight different persons. He found from 2.53 per cent to 13.54 per cent of the dry fecal substance made up of bacteria, with an average of 8.67 per cent, a value much lower than the result obtained by Strasburger. Steele[22] employed a modification of Strasburger's method. In three normal adults he found the bacterial substance to be 18.1, 21.1, and 18.2 per cent of the fecal dry substance, and the bacterial dry substance excreted daily, 7.44, 4.99, and 2.74 gm. or an average of 5.06 gm. per diem, comparatively close to the result obtained by Strasburger.

From this brief review of the more important literature it is evident that the results of the enumeration method do not agree closely with those of the gravimetric method, as the following extreme figures will indicate.

TABLE 1.

	Bacteria per Milligram Moist Feces	Daily Fecal Bacteria	Daily Bacterial Dry Substance Gm.	Bacteria in Fecal Dry Substance—Per ct.	Daily Bacterial Nitrogen Gm.
Klein	58,800,000	8.8×10^{12}	0.293	0.13	0.00439
Strasburger	....	128.0×10^{12}	8.100	33.00	0.83700

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* Klein[12] now regards these estimations as inaccurate and has repeated the calculations on a different basis, arriving at much higher results.

EXPERIMENTAL RESULTS.

In Table 2, A to L, are given the data of the quantitative examination of 204 stools from 12 different subjects. The bacteria were counted by the Winterberg method and by the Klein method for each of these. The tables also show the results of the gravimetric determination of the bacteria by the method of Strasburger upon 141 of the 204 stools. Of these 141 examinations 42 of the first are not considered sufficiently accurate for comparative purposes. The last 99 gravimetric determinations were performed according to the method we have detailed above (page 129), are nearly all done in duplicate, and have the nitrogen determination made upon the bacterial residues. From these the determination of Subject D, May 28, should be excluded on account of the low nitrogen result, leaving 98 determinations which we regard as fairly representative of the results to be expected from this method.

The highest single count by the Winterberg method occurs in Subject C, Nov. 26, 1907, 816×10^6 per milligram. Among the recent examinations the highest count occurs in Subject H, June 12, 1908, 751×10^6 bacteria per milligram fresh feces. The lowest count by this method occurs in Subject B, Nov. 10, 1907, 124×10^6 bacteria per milligram feces. The lowest counts among the recent examinations occur in Subject D, July 1, 176×10^6 per milligram, and Subject L, May 25, 1908, 220×10^6 per milligram. Of the 12 individuals, the highest average counts of all determinations by this method are in Subject C, 490×10^6 per milligram, and Subject H, 481×10^6 per milligram; the lowest average counts are in Subject B, 275×10^6 per milligram, Subject K, 280×10^6 per milligram, and Subject D, 302×10^6 per milligram. The grand average of all 12 subjects by this method of counting is 384×10^6 bacteria per milligram of fresh feces.

Enumeration by the Eberle-Klein method gave the highest single counts in Subject H, April 25, 1908, 723×10^6 per milligram, and in Subject H, June 12, 1908, 663×10^6 per milligram; the lowest single counts in Subject L, Jan. 20, 149×10^6 per milligram, and Subject B, Dec. 9, 1907, 163×10^6 per milligram. Of the 12 individuals, Subject H with 469×10^6 per milligram and Subject C with 432×10^6 per milligram show the highest average counts by this method. The lowest are in Subject K, 293×10^6, and Subject D, 309×10^6 per milligram. The grand average of the 12 subjects by this counting method is 375×10^6 bacteria per milligram feces. The results of these two methods of counting correspond fairly well, the differences being less than 30 per cent in most instances. Much larger discrepancies occur, for example in Subject L, May 25, but these are not common, and as a rule the average of the two counts would appear to be reliable within 20 per cent.

The mean of the two counts has been used as the basis of comparison with the gravimetric results, for the calculation of the number of bacteria per gram of fecal dry substance, and for the estimation of the daily excretion of bacteria. The highest average mean value occurs in Subject H, 475×10^6 per milligram, and the lowest in Subject K, 287×10^6 per milligram. The grand average mean of the two counts is 379×10^6 per milligram.

Of the 204 stools in which the bacteria were counted, the dry substance was determined in 148. For these the number of bacteria per gram dry fecal substance has been calculated and is recorded in Table 2, A to L. The lowest single value is in Subject D, Jan. 9, 983×10^9 bacteria per gram fecal dry substance. The highest single value is in Subject C, Feb. 20, 2642×10^9 bacteria per gram fecal dry substance. Subject D also shows the lowest average value of all examinations, 1371×10^9 bacteria per gram dry feces, and Subject C the highest average value, 1856×10^9 bacteria per gram dry feces. The grand average of all the examinations on the twelve individuals is 1587×10^9 bacteria per gram dry feces.

The quantitative determination of the fecal bacteria by the Strasburger method yielded results which are expressed first as amount of bacterial dry substance per gram moist feces, and second as percentage bacterial dry substance in fecal dry substance. The first set of figures is comparable with the results of enumeration and the second set is comparable with the values obtained by Strasburger and by Steele. These percentages of bacterial substance in dry substance have also been employed to calculate the daily bacterial excretion (Table 4, A to L).

The greatest quantity of bacterial dry substance per gram moist feces in a single determination was obtained in Subject H, July 24, 1908, 0.132 gm. The next highest value occurred in the same subject June 12, 0.127 gm. The smallest quantity was found in Subject

L, May 25, 0.026 gm.; the next smallest in Subject D, July 17, 0.028 gm. Of the 12 individuals, Subject H gave the highest average value for all examinations, 0.097 gm. dry bacteria per gm. moist feces; Subjects D and K gave the lowest, 0.041 gm. in each case. The grand average for the 12 individuals is 0.070 gm. dry bacteria per gram fresh feces.

The duplicate gravimetric determinations agree fairly well as a rule and in some cases the agreement is very close. Large discrepancies are not rare, however, for example Subject I, April 29, and Subject B, May 1. These are probably due to definite error in technic rather than inherent in the method. The determination in duplicate and the estimation of the nitrogen in the bacterial residues checked them in such a way that the average results of the gravimetric method are probably more reliable, as a rule, than the mean of the two enumerations.

The percentage of dry bacterial substance in the fecal dry substance for a single determination was highest in Subject H, July 24, 42.53 per cent. Other high values were found in Subject H, June 12, 40.17 per cent, and in Subject F, May 7, 40.03 per cent. The lowest values were found in Subject D, July 17, 13.26 per cent, Subject L, May 25, 14.03 per cent, and Subject E, March 16, 14.53 per cent.

Of the 12 individuals Subject H showed the highest average bacterial substance in the dry fecal substance, 34.62 per cent (mean of nine determinations); Subject I followed with 32.06 per cent. The lowest average values were in Subject K, 20.08 per cent, and Subject D, 21.10 per cent. The grand average for the 12 individuals is 26.89 per cent bacterial substance in the dry fecal substance.

The determinations of the nitrogen in the bacterial residues were made by Mr. Gill in the analytical division of the Laboratory at the suggestion of Professor Grindley. Residues in which the nitrogen fell below 9.5 per cent were considered too impure to be used as a basis of calculation. Nearly all of them show more than 10 per cent of nitrogen, a few as much as 12 per cent. The grand average of all the nitrogen determinations on the satisfactory residues is 10.96 per cent.

A comparison of the quantitative determination of the fecal bacteria by the counting method with the results of the gravimetric method is of some interest. The results of the two methods correspond to a cer-

TABLE 2.

Fecal Bacteria. Quantitative Determinations.

SUBJECT A.

Laboratory No.	Date	Wt. of Stool Gm.	Bacteria Per Gram by Enumeration $\times 10^9$						Dry Bacteria Per Gram by Strasburger Method			Dry Substance Per Gram.			Bacteria in Dry Feces		Per cent of Nitrogen in Bacteria		
			Winterberg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per gram $\times 10^9$	Per cent by Wt.	1	2	Mean
B 37	Nov. 12	63.2	266	..	406	..	336	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 60	Nov. 26	127.8	356	..	256	..	306	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 83	Dec. 10	115.4	371	..	344	..	358	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B110	Jan. 6	70.4	260	..	228	..	244	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B122	Jan. 22	80.5	377	34	302	27	339	30	0.112	.....	0.112	0.272	.....	0.272	1246	41.52	.....	.....	.....
B134	Feb. 6	118.7	380	32	317	27	348	29	0.118	.....	0.118	0.260	.....	0.260	1338	45.28	.....	.....	.....
B146	Feb. 24	126.6	363	27	332	25	348	26	0.134	0.137	0.135	0.276	.....	0.276	1261	49.09	7.58	7.47	7.53
B158	Mar. 11	4.1	414	66	479	76	447	71	0.063	.....	0.063	0.225	.....	0.225	1987	28.13	7.53	....	7.53
B170	Mar. 27	62.2	518	62	426	51	472	56	0.091	0.076	0.084	0.345	.....	0.345	1368	24.22	9.90	10.80	10.35
B182	Apr. 10	60.2	443	81	372	68	407	74	0.052	0.059	0.055	0.292	0.293	0.292	1394	18.97	12.49	11.26	11.88
B194	Apr. 27	34.9	477	51	476	51	478	51	0.095	0.091	0.093	0.354	0.355	0.354	1345	26.26	.....	10.82	10.82
B206	May 13	43.4	360	58	358	58	359	58	0.062	0.063	0.062	0.347	0.353	0.350	1026	17.81	.....	10.46	10.46
B218	May 28	18.0	471	46	449	44	460	45	0.103	(0.131)	0.103	0.332	.....	0.332	1386	35.34	11.55	(8.24)	11.55
B230	June 15	60.4	534	45	545	46	539	45	0.113	0.124	0.119	0.339	0.338	0.339	1590	35.00	10.40	10.16	10.28
B242	July 1	32.5	626	70	319	35	472	52	0.087	0.093	0.090	0.347	0.348	0.347	1360	25.88	10.30	10.12	10.21
B254	July 17	63.3	375	101	319	86	347	94	0.038	0.036	0.037	0.242	0.244	0.243	1428	15.32	10.34	10.46	10.40
B266	July 31	64.7	564	87	318	49	441	68	0.067	0.063	0.065	0.330	0.332	0.331	1332	19.65	.....	11.36	11.36
Average		.....	421	67*	367	54*	394	60*	.....	.....	0.079*	.....	.....	0.326*	1390	24.27*	.....	.....	10.81*
Minimum		.....	.....	45*	..	35*	..	45*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum		.....	.....	101*	....	86*	....	94*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT B.

Laboratory No.	Date	Wt. of Stool Gm.	Winterberg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per gram $\times 10^9$	Per cent by Wt.	1	2	Mean
B 33	Nov. 10	129.9	124	..	305	..	215	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 56	Nov. 22	101.8	206	..	321	..	309	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 79	Dec. 9	130.9	282	..	163	..	223	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B102	Dec. 26	166.7	136	..	166	..	151	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B114	Jan. 12	123.4	192	..	304	..	248	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B126	Jan. 27	99.1	276	39	365	52	325	46	0.070	.....	0.070	0.207	.....	0.207	1570	34.04	.....	.....	.....
B138	Feb. 13	157.8	220	28	257	32	239	30	0.082	0.078	0.080	0.172	.....	0.172	1390	46.27	7.25	7.40	7.32
B150	Mar. 2	44.6	248	46	320	59	284	53	0.055	0.053	0.054	0.190	.....	0.190	1495	28.45	10.28	10.36	10.32

B162	Mar. 16	82.5	248	56	343	78	296	67	0.043	0.044	0.044	0.165	.....	0.165	1794	26.56	10.55	10.44	10.50
B174	Apr. 1	37.2	368	..	364	..	366	..	.....	.....	.....	(0.224	.....	0.224	1634	.....	.....	.....	.....
B186	Apr. 16	36.8	396	73	436	81	416	77	0.053	0.055	0.054	0.279	0.278	0.278	1496	19.55	10.48	10.38	10.43
B198	May 1	22.1	392	75	365	72	379	73	0.037	0.067	0.052	0.255	0.264	0.260	1458	19.98	10.95	.....	10.95
B210	May 18	94.7	252	70	340	95	296	82	0.041	0.031	0.036	0.191	0.180	0.185	1600	19.56	.....	.....	.....
B222	June 3	184.2	207	49	389	93	298	71	0.046	0.038	0.042	0.155	0.153	0.154	1935	27.08	10.96	12.84	11.90
B234	June 19	128.3	324	70	436	95	380	83	0.043	0.049	0.046	0.204	0.184	0.194	1959	23.80	11.17	11.58	11.38
B246	July 6	47.3	444	63	405	58	425	61	0.070	0.070	0.070	0.254	0.251	0.252	1683	27.78	11.00	11.18	11.09
B258	July 22	9.9	276	73	262	69	269	71	0.038	0.038	0.038	0.179	.....	0.179	1503	21.27	10.54	(6.42)	10.54
Average.....		.....	275	66*	326	80*	301	73*	.....	.....	0.048*	.....	.....	0.208*	1626	23.20*	.....	.....	10.97*
Minimum...		.....	.....	49*	..	58*	..	61*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum..		.....	.....	75*	..	95*	..	83*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT C.

B 39	Nov. 13	50.9	321	..	360	..	341	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 62	Nov. 26	27.5	816	..	530	..	673	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 85	Dec. 12	65.0	546	..	330	..	438	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B104	Dec. 28	56.5	377	..	342	..	359	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B116	Jan. 14	105.8	382	..	427	..	405	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B128	Jan. 29	65.8	398	..	406	..	402	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B140	Feb. 20	27.2	494	53	420	45	457	49	0.096	0.091	0.094	0.173?	.....	0.173?	2642	53.98?	10.48	10.86	10.67
B154	Mar. 6	81.2	484	77	442	70	463	73	0.064	0.062	0.063	0.201	.....	0.201	2303	31.34	10.87	11.25	11.06
B166	Mar. 23	18.9	454	38	418	35	436	37	0.114	0.122	0.118	0.304	.....	0.304	1434	38.84	11.12	11.41	11.26
B178	Apr. 6	54.2	414	68	336	55	375	61	0.059	0.062	0.061	0.224	0.231	0.227	1652	26.76	11.53	12.11	11.82
B190	Apr. 22	53.5	435	68	488	76	461	72	0.050	0.079	0.064	0.236	0.250	0.243	1897	26.48	11.36	10.96	11.16
B202	May 7	17.9	526	49	596	56	561	52	0.107	0.107	0.107	0.317	0.312	0.315	1781	33.99	.....	11.17	11.17
B214	May 23	46.5	517	63	397	48	457	56	0.075	0.090	0.082	0.298	0.291	0.294	1554	27.98	11.33	11.37	11.35
B226	June 8	65.2	451	56	428	53	439	54	0.081	.....	0.081	0.269	0.271	0.270	1626	29.86	.....	.....	.....
B238	June 25	21 4	720	80	637	71	678	75	0.095	0.084	0.090	0.297	0.330	0.313	2166	28.56	11.48	11.56	11.52
B250	July 13	42.3	540	58	458	49	499	54	0.093	0.094	0.093	0.290	0.299	0.295	1692	31.62	11.20	10.97	11.09
B262	July 27	85.4	454	69	323	49	388	59	0.065	0.067	0.066	0.233	0.234	0.233	1665	28.41	.....	11.00	11.00
Average.....		.....	490	61*	432	55*	461	58*	.....	.....	0.085*	.....	.....	0.277*	1856	30.28*	.....	.....	11.30*
Minimum...		.....	..	38*	..	35*	..	37*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum..		.....	..	80*	..	76*	..	75*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT D.

B 34	Nov. 11	115.6	365	..	332	..	348	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 57	Nov. 23	222.3	518	..	376	..	448	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

* Determinations previous to March 16 are not included in this calculation.

TABLE 2.—*Continued*

SUBJECT D

Labor-atory No.	Date	Wt. of Stool Gm.	Bacteria per Gram by Enumeration × 10⁹						Dry Bacteria per Gram by Strasburger Method			Dry Substance per Gram			Bacteria in Dry Feces		Per cent of Nitrogen in Bacteria		
			Winter-berg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per gram × 10⁹	Per cent by Wt.	1	2	Mean
B 80	Dec. 10	128.9	272	..	307	..	289	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 99	Dec. 23	20.0	336	..	439	..	387	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B112	Jan. 9	139.0	220	32	236	35	228	34	0.068	.....	0.068	0.232	.....	0.232	983	29.48	.....	.....	.....
B123	Jan. 25	168.6	284	23	240	20	262	22	0.121	.....	0.121	0.264	.....	0.264	992	45.88	.....	.....	.....
B135	Feb. 10	49.0	300	33	276	30	288	31	0.092	.....	0.092	0.205	.....	0.205	1398	44.77	9.90	.....	9.90
B147	Feb. 27	64.8	276	45	270	44	270	44	0.049	0.062	0.062	0.240	.....	0.240	1125	25.91	10.30	10.43	10.37
B157	Mar. 11	73.7	316	72	389	88	352	80	0.047	0.041	0.044	0.249	0.251	0.250	1408	17.52	10.94	.....	10.94
B169	Mar. 27	123.8	380	78	264	54	322	66	0.052	0.047	0.049	0.202	.....	0.202	1594	24.28	12.26	11.76	12.01
B181	Apr. 10	81.7	284	65	332	75	308	70	0.045	0.043	0.044	0.200	0.202	0.201	1532	21.81	12.50	12.67	12.58
B193	Apr. 27	235.6	224	53	280	67	252	60	0.040	0.045	0.042	0.163	0.197	0.180	1400	23.35	10.92	11.29	11.10
B205	May 13	157.3	272	57	385	80	328	68	0.061	0.036	0.048	0.190	0.214	0.202	1624	24.00	10.40	10.82	10.61
B217	May 28	29.6	356	(52†)	321	(47†)	339	(49†)	(0.066	0.073	0.069†)	0.189	0.208	0.198	1662	34.95†	8.96	7.95	8.45†
B220	June 15	45.0	276	59	254	54	265	56	0.057	0.038	0.047	0.187	0.187	0.187	1417	25.27	11.14	11.23	11.18
B241	July 1	96.1	176	50	210	60	193	55	0.037	0.034	0.035	0.173	0.168	0.170	1135	20.75	11.32	9.44	10.38
B253	July 17	22.0	288	103	325	116	307	110	0.027	0.029	0.028	0.224	0.193	0.208	1471	13.26	11.73	11.14	11.43
B265	July 31	171.7	288	85	320	94	304	89	0.032	0.036	0.034	0.213	0.206	0.210	1448	16.08	11.49	11.46	11.47
Average.....		.....	302	69*	309	75*	305	72*	.....	.....	0.041*	.....	.....	0.195*	1371	21.10*	.....	.....	11.35*
Minimum...		.....	....	50*	..	54*	..	55*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	103*	..	116*	..	110*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT E.

Labor-atory No.	Date	Wt. of Stool Gm.	Winter-berg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per gram × 10⁹	Per cent by Wt.	1	2	Mean
B 41	Nov. 14	40.0	248	..	260	..	254	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 64	Nov. 27	160.8	256	..	303	..	279	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 87	Dec. 13	118.5	323	..	406	..	365	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B106	Dec. 30	24.8	516	..	346	..	431	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B118	Jan. 16	73.4	444	..	465	..	454	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B130	Jan. 31	46.9	622	38	365	23	494	30	0.162	.....	0.162	0.346	.....	0.346	1428	46.87	.....	.....	.....
B142	Feb. 19	126.2	398	39	399	40	398	39	0.097	0.105	0.101	0.231	.....	0.231	1723	43.88	10.10	9.73	9.96
B149	Mar. 2	88.2	478	59	483	60	480	59	0.078	0.085	0.081	0.273	.....	0.273	1758	29.72	10.50	10.58	10.54
B161	Mar. 16	22.5	276	99	322	115	299	107	0.027	0.028	0.028	0.190	.....	0.190	1574	14.53	10.63	9.82	10.23
B173	Apr. 1	24.7	510	..	560	..	535	..	.....	.....	.....	0.266	.....	0.266	2011	.....	.....	.....	.....

B185	Apr. 16	32.5	468	51	413	45	441	48	0.095	0.088	0.092	0.285	0.288	0.286	1542	32.02	11.17	11.46	11.31
B197	May 1	40.5	533	68	495	63	514	66	0.085	0.071	0.078	0.327	0.321	0.324	1586	24.04	10.36	10.89	10.63
B209	May 18	18.5	658	89	368	50	513	69	0.074	.....	0.074	0.341	.....	0.341	1504	21.56	10.13	.....	10.13
B221	June 3	138.8	333	46	354	48	343	47	0.077	0.070	0.073	0.231	0.233	0.232	1478	31.71	11.39	11.37	11.38
B233	June 19	53.0	456	63	479	67	468.	65	0.071	0.072	0.072	0.289	0.272	0.281	1665	25.59	11.68	11.51	11.60
B245	July 6	33.5	397	55	349	48	373	52	0.071	0.073	0.072	0.240	0.231	0.236	1581	30.42	11.17	11.64	11.40
B257	July 22	63.9	658	87	481	63	569	75	0.076	.....	0.076	0.291	0.284	0.288	1976	26.46	10.80	.....	10.80
Average.....			446	70*	403	62*	424	66*	.....	.....	0.071*	.....	.....	0.272*	1652	25.79*	.....	.....	10.94*
Minimum...		.....	....	46*	...	45*	..	47*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	...	99*	...	115*	..	107*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT F.

B 38	Nov. 13	48.8	400	..	393	..	396	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 61	Nov. 26	44.0	364	..	496	..	430	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 84	Dec. 12	25.9	328	..	393	..	360	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B103	Dec. 28	21.5	464	..	455	..	459	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B115	Jan. 14	9.0	464	..	466	..	465	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B127	Jan. 29	39.2	436	31	331	24	384	28	0.139	.....	0.139	0.318	.....	0.318	1208	.....	.....	.....	.....
B139	Feb. 17	69.8	522	34	415	27	469	33	0.155	.....	0.155	0.388	.....	0.388	1209	39.77	9.18	.....	9.18
B153	Mar. 6	79.4	232	31	294	40	263	36	0.042	0.074	0.074	0.214	.....	0.214	1229	34.42	12.86	10.62	10.62
B165	Mar. 23	13.9	460	39	453	39	456	39	0.122	0.112	0.117	0.311	.....	0.311	1466	37.71	10.89	10.73	10.81
B177	Apr. 6	32.5	444	63	486	68	465	66	0.073	0.069	0.071	0.252	0.254	0.253	1838	28.12	11.44	12.33	11.88
B189	Apr. 22	42.7	476	55	521	61	498	58	0.084	0.088	0.086	0.296	0.294	0.295	1688	29.16	11.20	10.55	10.88
B201	May 7	22.8	492	40	461	38	476	39	0.128	0.116	0.122	0.306	0.303	0.304	1566	40.03	.....	10.39	10.39
B213	May 23	54.5	412	57	404	56	408	57	0.066	0.078	0.072	0.253	0.259	0.255	1600	28.14	10.23	.....	10.23
B225	June 8	57.9	444	62	511	71	477	66	0.072	0.072	0.072	0.287	0.284	0.286	1647	25.22	10.42	10.08	10.25
B237	June 25	23.1	460	65	386	54	423	60	0.073	0.070	0.071	0.304	0.305	0.304	1391	23.42	11.16	11.35	11.25
B249	July 13	35.3	472	68	365	53	418	61	0.070	0.069	0.069	0.250	0.248	0.249	1679	27.83	11.30	11.25	11.27
B261	July 27	83.4	340	83	456	111	398	97	0.042	0.040	0.041	0.206	0.211	0.200	1904	19.58	10.72	11.42	11.07
Average.....		.....	401	59*	405	61*	403	60*	.....	.....	0.080*	.....	.....	0.274*	1535	28.80*	.....	.....	10.89*
Minimum...		.....	....	39*	..	38*	..	39*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	83*	..	111*	..	97*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT G

B 42	Nov. 15	137.5	224	..	193	..	208	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 65	Nov. 29	46.7	380	..	302	..	341	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 88	Dec. 14	128.4	166	..	192	..	179	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

* Determinations previous to March 16 are not included in this calculation.

† Not included in the average calculation on account of low nitrogen.

TABLE 2.—*Continued*

SUBJECT G

Laboratory No.	Date	Wt. of Stool Gm.	Bacteria per Gram by Enumeration $\times 10^9$						Dry Bacteria per Gram by Strasburger Method			Dry Substance per Gram			Bacteria in Dry Feces		Per Cent of Nitrogen in Bacteria		
			Winterberg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per gram $\times 10^9$	Per cent by Wt.	1	2	Mean
B107	Jan. 2	114.3	298	..	356	..	327	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B119	Jan. 20	26.9	260	19	259	19	260	19	0.140	.....	0.140	0.252	.....	0.252	1032	55.56	.....	.....	.....
B131	Feb. 3	38.8	378	24	380	24	379	24	0.157	.....	0.157	0.305	.....	0.305	1243	51.72	.....	.....	.....
B143	Feb. 21	66.9	593	46	475	37	534	41	0.116	0.143	0.130	0.299	.....	0.299	1786	43.44	10.01	9.61	9.81
B152	Mar. 4	41.0	518	46	441	39	480	43	0.111	0.114	0.112	0.304	.....	0.304	1579	36.96	.....	9.93	9.93
B164	Mar. 19	73.4	527	53	351	35	439	44	0.094	0.107	0.100	0.293	0.291	0.292	1503	34.37	10.55	.....	10.55
B176	Apr. 3	31.4	527	..	421	..	474	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B188	Apr. 20	59.8	496	65	439	58	468	62	0.076	0.076	0.076	0.284	0.274	0.279	1677	27.23	10.63	10.95	10.79
B200	May 4	39.5	408	42	483	49	446	46	0.098	.....	0.098	0.296	0.297	0.297	1522	33.02	.....	10.72	10.72
B212	May 20	39.5	600	57	393	37	497	47	0.100	0.109	0.105	0.294	0.305	0.299	1662	34.99	11.18	10.27	10.72
B224	June 5	229.5	285	77	294	79	290	78	0.037	0.036	0.037	0.167	0.154	0.160	1812	22.81	11.64	11.69	11.67
B236	June 22	229.5	382	71	450	83	416	77	0.052	0.055	0.054	0.193	0.197	0.195	2133	28.85	11.78	(14.80)	11.78
B248	July 9	97.6	526	60	440	50	483	55	0.092	0.084	0.088	0.309	0.315	0.312	1548	28.09	10.62	10.66	10.64
B264	July 29	40.0	688	78	478	54	583	66	0.087	0.089	0.088	0.320	0.312	0.316	1845	27.82	10.96	11.04	11.00
Average.....		.....	427	63*	373	56*	400	59*	.....	.....	0.081*	.....	.....	0.269*	1612	27.15*	.....	.....	10.98*
Minimum...		.....	....	42*	..	35*	..	44*	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
Maximum...		.....	....	78*	..	83*	..	78*	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....	.....
SUBJECT H																			
B 48	Nov. 18	79.0	399	..	355	..	377	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 71	Dec. 3	31.8	524	..	546	..	535	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 94	Dec. 19	102.1	384	..	344	..	364	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B109	Jan. 6	53.8	456	..	369	..	412	..	.....	.....	.....	0.191	.....	0.191	2157	.....	.....	.....	.....
B121	Jan. 22	38.2	420	26	347	22	383	24	0.159	.....	0.159	0.331	.....	0.331	1157	48.03	.....	.....	.....
B133	Feb. 6	21.0	396	38	332	32	364	35	0.103	.....	0.103	0.230	.....	0.230	1583	44.92	.....	.....	.....
B145	Feb. 24	70.7	404	27	497	33	451	30	0.150	(0.176)	0.150	0.274	.....	0.274	1646	54.71	8.89	7.81	8.39
B156	Mar. 10	27.7	517	49	518	49	517	49	0.100	0.109	0.105	0.279	.....	0.279	1853	37.51	11.37	11.08	11.23
B168	Mar. 25	155.9	421	44	484	50	453	47	0.088	0.104	0.096	0.255	0.251	0.253	1791	37.95	11.44	11.49	11.47
B180	Apr. 8	63.5	532	53	468	47	500	50	0.099	0.101	0.100	0.328	0.320	0.324	1543	30.74	11.91	11.08	11.49
B192	Apr. 25	79.9	570	48	723	61	647	54	0.114	0.123	0.119	0.304	0.311	0.307	2107	38.64	11.12	(6.04)	11.12
B204	May 11	22.1	444	46	468	49	456	48	0.081	0.110	0.096	0.316	0.307	0.311	1466	30.70	10.90	10.57	10.73
B216	May 25	80.9	439	55	584	73	512	64	0.075	0.084	0.080	0.272	0.268	0.270	1896	29.44	11.34	11.17	11.25

B228	June 12	20.6	751	59	663	52	707	56	0.126	0.129	0.127	0.320	0.314	0.317	2230	40.17	11.17	10.74	10.96
B240	June 29	109.4	559	75	513	68	536	71	0.069	0.082	0.075	0.242	0.237	0.239	2243	31.51	11.76	10.89	11.32
B252	July 15	129.5	320	64	324	65	322	64	0.050	0.050	0.050	0.170	0.168	0.169	1905	29.86	11.63	11.68	11.65
B260	July 24	23.6	647	49	432	33	540	41	0.134	0.131	0.132	0.310	0.312	0.311	1736	42.53	11.47	11.44	11.45
Average.....		.....	481	55*	469	55*	475	55*	.....	.....	0.097*	.....	.....	0.278*	1808	34.62*	.....	.....	11.27*
Minimum...		.....	....	44*	..	33*	..	41*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	75*	..	73*	..	71*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT I

B 32	Nov. 11	124.7	260	..	241	..	251	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 55	Nov. 22	69.0	394	..	303	..	348	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 78	Dec. 9	136.0	248	..	333	..	290	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B101	Dec. 26	216.8	331	..	250	..	291	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B113	Jan. 12	73.8	321	..	348	..	335	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B125	Jan. 27	153.6	314	37	266	31	290	34	0.086	.....	0.086	0.207	.....	0.207	1401	41.79	.....	.....	.....
B137	Feb. 14	69.6	454	37	542	44	498	40	0.120	0.128	0.124	0.281	.....	0.281	1772	44.09	9.50	8.89	9.20
B160	Mar. 14	237.8	296	64	268	58	282	61	0.044	0.048	0.046	0.189	0.193	0.191	1476	24.03	10.43	10.06	10.24
B172	Mar. 31	107.0	416	..	468	..	442	..	.....	.....	.....	0.260	.....	0.260	1693	.....	.....	.....	.....
B184	Apr. 13	128.6	308	33	490	52	399	43	0.075	0.111	0.093	0.264	0.255	0.259	1541	35.68	11.61	11.06	11.33
B196	Apr. 29	80.0	480	64	434	58	457	61	0.075	0.040	0.075	0.312	0.312	0.312	1465	24.00	10.83	11.05	10.94
B208	May 15	88.0	424	46	533	58	478	52	0.090	0.093	0.092	0.248	0.246	0.247	1935	37.29	.....	10.17	10.17
B220	June 1	186.5	392	44	403	45	397	44	0.089	0.090	0.090	0.252	0.250	0.251	1582	35.78	11.06	10.47	10.76
B232	June 17	112.3	480	..	505	..	492	..	.....	.....	.....	0.285	0.277	0.281	1751	.....	.....	.....	.....
B244	July 3	125.1	588	75	636	82	612	78	0.081	0.076	0.078	0.266	0.278	0.272	2250	28.80	10.90	10.46	10.68
B256	July 20	55.5	584	70	568	68	576	69	0.085	0.082	0.083	0.280	0.258	0.270	2133	30.82	11.60	11.44	11.52
Average.....		.....	393	55*	412	61*	402	58*	.....	.....	0.085*	.....	.....	0.268*	1727	32.06*	.....	.....	10.90*
Minimum...		.....	....	33*	..	45*	..	43*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	75*	..	82*	..	78*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT J

B 31	Nov. 9	124.2	131	..	184	..	158	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 54	Nov. 21	115.0	297	..	347	..	322	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 77	Dec. 7	137.7	284	..	274	..	279	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B100	Dec. 23	17.7	290	..	245	..	268	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B111	Jan. 9	65.7	250	32	346	44	298	38	0.078	.....	0.078	0.272	.....	0.272	1096	45.35	.....	.....	.....
B124	Jan. 25	126.6	382	29	237	18	309	23	0.132	.....	0.132	0.274	.....	0.274	1128	48.18	.....	.....	.....
B136	Feb. 10	107.6	210	30	230	32	220	31	0.071	.....	0.071	0.158	.....	0.158	1392	44.57	10.06	.....	10.06
B148	Feb. 27	60.8	389	29	383	29	386	29	(0.108)	0.133	0.133	0.293	.....	0.293	1317	45.47	9.25	8.87	9.06
B159	Mar. 13	33.1	347	44	380	48	364	46	0.081	0.077	0.079	0.286	0.277	0.282	1291	28.05	9.47	10.50	9.98

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* Determinations previous to March 16 are not included in this calculation.

TABLE 2.—*Continued*

SUBJECT J

Labor-atory No.	Date	Wt. of Stool Gm.	Bacteria per Gram by Enumeration × 10^9						Dry Bacteria per Gram by Strasburger Method			Dry Substance per Gram			Bacteria in Dry Feces		Per Cent of Nitrogen in Bacteria		
			Winter-berg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per Gram × 10^9	Per Cent by Wt.	1	2	Mean
B171	Mar. 30	78.4	365	37	438	44	401	40	0.098	0.103	0.100	0.290	0.294	0.292	1373	34.38	10.29	10.02	10.15
B183	Apr. 14	49.8	428	50	450	53	439	52	0.086	0.083	0.085	0.343	0.338	0.341	1287	26.79	10.44	10.93	10.68
B195	Apr. 29	80.1	325	40	280	35	302	37	0.088	0.075	0.081	0.241	0.225	0.232	1302	34.98	.....	10.71	10.71
B207	May 15	48.2	366	42	400	46	383	44	0.081	0.093	0.087	0.224	0.224	0.224	1710	38.81	(8.52)	10.43	10.43
B219	June 1	87.0	436	48	515	57	476	53	0.091	0.090	0.090	0.266	0.257	0.262	1817	34.49	10.26	.....	10.26
B231	June 17	94.6	377	..	360	..	368	..	.....	.....	.....	0.239	0.256	0.248	1484	.....	.....	.....	.....
B243	July 4	113.9	379	76	366	73	372	74	0.049	0.051	0.050	0.219	0.209	0.214	1738	23.43	10.52	10.40	10.46
B255	July 20	62.5	276	89	179	58	228	74	0.029	0.033	0.031	0.171	0.151	0.161	1416	19.24	11.03	10.57	10.80
Average.....		.....	325	55*	330	52*	328	53*	.....	.....	0.075*	.....	.....	0.247*	1412	30.30*	.....	.....	10.50*
Minimum...		.....	....	37*	..	35*	..	37*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	89*	..	73*	..	74*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT K

Labor-atory No.	Date	Wt. of Stool Gm.	Winter-berg	Ratio	Klein	Ratio	Mean	Ratio	1	2	Mean	1	2	Mean	No. per Gram × 10^9	Per Cent by Wt.	1	2	Mean
B 40	Nov. 14	39.0	300	..	327	..	313	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 63	Nov. 27	101.3	258	..	259	..	259	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 86	Dec. 13	20.6	264	..	200	..	232	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B105	Dec. 30	104.2	272	..	182	..	227	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B117	Jan. 16	78.1	212	22	317	33	264	27	0.097	.....	0.097	.....	.....	.....	....	.....	.....	.....	.....
B129	Jan. 31	24.0	324	36	277	30	300	33	0.091	.....	0.091	0.226	.....	0.226	1327	40.02	.....	.....	.....
B141	Feb. 19	37.9	192	30	203	32	198	31	0.059	0.069	0.064	0.175	.....	0.175	1131	36.67	9.36.	9.51	9.44
B151	Mar. 4	49.0	384	65	385	65	385	65	0.057	0.061	0.059	0.256	.....	0.256	1504	23.18	10.95	11.35	11.15
B163	Mar. 19	76.7	320	89	397	110	358	99	0.038	0.033	0.036	0.185	0.189	0.187	1914	19.03	10.20	10.53	10.36
B175	Apr. 3	104.4	252	..	312	..	282	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B187	Apr. 20	36.7	322	47	390	57	356	52	0.077	0.061	0.069	0.235	0.250	0.243	1465	28.43	10.39	10.58	10.48
B199	May 4	37.0	256	71	264	73	260	72	0.036	0.037	0.036	0.210	0.208	0.209	1244	17.41	10.97	9.77	10.37
B211	May 20	56.2	224	75	328	109	276	95	0.029	0.031	0.030	0.191	0.202	0.197	1401	15.34	11.23	11.12	11.17
B223	June 5	49.2	324	85	309	81	317	83	0.043	0.033	0.038	0.244	0.245	0.245	1294	15.44	10.71	10.95	10.83
B235	June 23	56.8	404	75	337	62	370	69	0.051	0.057	0.054	0.251	0.246	0.248	1492	21.70	10.74	9.21	9.97
B247	July 9	71.0	224	68	223	68	224	68	0.035	0.031	0.033	0.151	0.144	0.148	1514	22.46	11.34	9.13	10.24
B263	July 29	127.7	236	72	268	81	252	76	0.032	0.035	0.033	0.157	0.165	0.161	1565	20.79	12.11	11.17	11.64
Average.....		.....	280	73*	293	80*	287	77*	.....	.....	0.041*	.....	.....	0.205*	1441	20.08*	.....	.....	10.63*
Minimum...		.....	....	47*	..	57*	..	52*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	89*	..	110*	..	99*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

SUBJECT L

B 51	Nov. 19	73.3	494	..	407	..	451	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 74	Dec. 5	67.4	468	..	494	..	481	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B 97	Dec. 20	55.0	329	..	345	..	337	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B108	Jan. 2	27.5	335	..	326	..	331	..	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
B120	Jan. 20	120.4	201	32	149	24	175	28	0.063	.....	0.063	0.159	.....	0.159	1101	39.59	.....	.....	.....
B132	Feb. 3	23.8	204	30	203	29	203	29	0.069	.....	0.069	0.160	.....	0.160	1269	43.30	.....	.....	.....
B144	Feb. 21	83.2	360	30	379	32	370	31	0.111	0.130	0.120	0.278	.....	0.278	1331	43.24	9.98	9.68	9.83
B155	Mar. 9	12.3	508	46	451	41	479	44	0.110	.....	0.110	0.386	.....	0.386	1241	28.56	11.25	.....	11.25
B167	Mar. 25	85.7	308	68	372	83	340	76	0.043	0.047	0.045	0.188	0.201	0.195	1744	23.12	.....	10.18	10.18
B179	Apr. 8	54.3	352	53	428	65	390	59	0.066	0.067	0.066	0.246	0.221	0.233	1674	28.42	12.13	11.24	11.69
B191	Apr. 24	165.4	308	75	398	97	353	86	0.041	0.041	0.041	0.215	0.202	0.209	1689	19.79	11.46	11.28	11.37
B203	May 11	76.3	340	63	394	73	367	68	0.054	.....	0.054	0.225	.....	0.225	1631	24.20	.....	.....	.....
B215	May 25	17.5	220	85	384	148	302	116	0.023	0.029	0.026	0.182	.....	0.182	1659	14.03	12.15	11.50	11.82
B227	June 12	137.1	388	52	505	67	446	59	0.073	0.077	0.075	0.214	0.203	0.209	2134	36.14	(7.93)	10.98	10.98
B239	June 29	76.6	384	69	299	53	341	61	0.058	0.054	0.056	0.210	0.202	0.206	1655	27.31	11.39	10.90	11.15
B251	July 15	69.8	440	64	419	61	430	62	0.068	0.070	0.069	0.220	0.226	0.223	1928	30.88	10.94	10.96	10.95
B259	July 25	103.2	524	95	491	89	507	92	0.059	0.050	0.055	0.262	0.249	0.256	1980	21.45	10.55	10.85	10.70
Average.....		.....	363	69*	379	82*	371	75*	.....	.....	0.054*	.....	.....	0.215*	1618	25.04*	.....	.....	11.11*
Minimum...		.....	....	52*	..	53*	..	59*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....
Maximum...		.....	....	95*	..	148*	..	116*	.....	.....	.....	.....	.....	.....	....	.....	.....	.....	.....

* Determinations previous to March 16 are not included in this calculation.

tain extent. After each column of enumeration data in Table 2 has been inserted a column of figures headed "ratio," in which the relation between the two values is expressed. The figure given in this column multiplied by 10^{11} represents the number of bacterial cells according to that particular count, corresponding to one gram of bacterial dry substance according to the gravimetric analysis of the same sample of feces. Thus in Table 2, Subject A, May 13, 359×10^9 bacteria per gram moist feces correspond to 0.062 gm. of bacterial dry substance per gram moist feces. At this rate 58×10^{11} bacteria would yield one gram bacterial dry substance. Considering the mean data from March 16 to July 31 only, this ratio or factor is seen to vary from 116 in Subject L, May 25, and 110 in Subject D, July 17, to 37 in Subject C, March 23, 39 in Subject F, March 23, and 40 in Subject J, March 30. All intermediate values are also represented. Of the 12 individuals Subject K shows the highest average value for this ratio, namely, 77; Subject J shows the lowest, 53. The grand average of all the examinations is 64. According to these figures it would appear that in one case three times as many fecal bacteria are required to yield a gram of dry substance as in another instance, while the average lies about midway between the two extremes. The discrepancy is considerable but appears quite insignificant in comparison with the disagreement of the results of Klein and of Strasburger previously mentioned (p. 134).

In attempting to account for this variation, experimental error is the first factor which occurs to one. This certainly applies to some instances at least, as for example in Subject L, May 25, where the two counts do not agree and the gravimetric duplicates show a variation of 20 per cent. This seems not to be true in every case, however. The differences between the average ratio for the different individuals can hardly be explained in this way.

In our comparisons between the gravimetric and the counting methods we have assumed that the water content of the bacterial cells in the fecal suspension is constant. This, as a matter of fact, is not the case, as Cramer[1] and Lyons[16] have shown that bacteria do not possess a typical water content, but that it varies according to the conditions of growth, such as for example, the kind of medium[2] and age of the culture. Cramer[3] found that a culture of a single species shows wide variations in this respect. The bacteria of the feces include many species; there is considerable variation in the size of the various cells present and a very large majority of them are dead before the stool is passed. Spores are at times present in considerable numbers and these contain much more dry substance than vegetative cells.[1] One should therefore expect considerable variation in the chemical composition of the mixed fecal flora, especially in its dry-substance content. This variation in the dry-substance content of the bacterial cell is an important factor in accounting for the variation in the ratio between the two quantitative methods. By reference to Table 2, it will be seen that Subjects K, B, and D, the three subjects showing the lowest average counts, also show the smallest quantity of bacterial dry substance, the least percentage of dry substance in the feces, the least percentage of bacterial substance in the fecal dry substance, and **the highest enumeration-weight ratios,** namely, 77, 73, and 72. On the other hand, Subjects H, C, and I show high values for the counts and the gravimetric results, and the enumeration-weight ratio for each of these is low, 55, 58, and 58 respectively. Subject J with the lowest ratio is an exception to this general rule. In each individual also it will be seen that, as a general rule, an increase in the quantity of bacteria and an increase in the dry substance of the feces, is associated with a decrease in the enumeration-weight ratio of the bacteria. It would seem proper therefore to conclude that these various factors are to a certain extent associated with each other as cause and effect or as effects of the same cause.

Table 3 gives the mean of all determinations and the maximum and minimum single determinations of Table 2.

TABLE 3.

SUMMARY OF SINGLE EXAMINATIONS.

	BACTERIA COUNTED		DRY BACTERIAL SUBSTANCE		DRY SUBSTANCE IN FECES	ENUMERATION-WEIGHT RATIO
	Per Gram Fresh Feces	Per Gram Dry Feces	Per Gram Fresh Feces Grams	In Fecal Dry Substance Per cent	Per cent	
Mean............	375×10^9	1587×10^9	0.070	26.89	25.3	$64 \times 10^{11}:1$
Maximum........	816×10^9	2642×10^9	0.132	42.53	35.4	$116 \times 10^{11}:1$
Minimum........	124×10^9	983×10^9	0.026	14.03	16.1	$37 \times 10^{11}:1$

As has been said, the nitrogen in the dry bacterial residues was found to be from 10 to 12 per cent, with an average of 10.96 per cent. These figures agree with those of Strasburger.[23] Cramer,[2] in his analysis of various bacteria grown in pure culture on agar, found 10 to 13 per cent of nitrogen in the dry substance. Attention should, however, be called to the circumstance that his percentages were calculated on the total dry bacterial substance, while ours, and also those of Strasburger, are calculated upon the residue after extraction with alcohol and ether. When Cramer's results are calculated on extracted residue the nitrogen values become 10 to 15 per cent greater. Although it is not impossible that the alcohol and ether extraction removes nitrogenous bodies, yet we should expect the net result to be an increase in the nitrogen percentage of the residue. In pure cultures of *B. coli* grown upon agar, Leach[14] found 10.65 per cent nitrogen in an air-dried sample not at constant weight. In another determination in which 173 gm. of air-dried cell substance was analyzed, she obtained 11 per cent of nitrogen. The cell substance employed for these analyses was the residue after extraction with alcohol and ether in each case.

We have performed a few experiments in a small way to determine, upon pure cultures, the number of bacteria required to yield a gram of dry bacterial substance extracted as in Strasburger's method, and to determine the nitrogen content of such a residue. For this purpose we used pure cultures of *B. coli* grown upon agar in large flat bottles at 37° C. After from five to eight days the bacterial growth was carefully washed off with a small amount of 0.8 per cent salt solution and obtained in a concentrated suspension. This was thoroughly mixed. Dilute suspensions were then prepared and the number of bacteria estimated by the same methods as have been employed in counting the fecal bacteria. In addition to this, portions (4 to 5 c.c.) of the concentrated suspension were accurately wieghed in centrifuge tubes and suspended in 0.5 per cent hydrochloric acid, centrifugated, extracted, and dried, precisely as in the gravimetric method for separation of bacteria from feces. In three experiments of this kind, the relation between the two quantitative determinations agreed fairly well. In the first experiment the flask cultures were five days old and here 55×10^{11} bacteria by the counting methods corresponded to a gram of bacterial dry substance obtained by the gravimetric method. In the second experiment, in which the cultures were again five days old, the ratio was 46×10^{11} by count to a gram dry substance, and in the third experiment, with cultures eight days old, 57×10^{11} bacteria by count were found to be equivalent to a gram of the bacterial dry substance obtained by the gravimetric method. The nitrogen in these bacterial residues was 11.52 per cent in the first, 11.71 per cent in the second, and 11.80 per cent in the third experiment. It is thus evident that these methods yield, in their application to pure cultures of bacteria, results somewhat similar to those obtained upon bacteria of the feces. This we regard as important evidence of their reliability.

AVERAGE DAILY EXCRETION OF BACTERIA.

The average daily excretion of bacteria, of bacterial nitrogen, and the percentage of fecal nitrogen represented by the latter, were calculated for each subject, by applying the values obtained in the bacteriological examination of a single stool to the average daily quantity of feces, of fecal dry substance, and of nitrogen, for the eight-day period in which the bacteriological examination occurred. The principal objection tot his calculation lies in the circumstance that the stool examined was not necessarily a fair sample of the feces of that period; but in a series of examinations, errors introduced by this chance would tend to equalize each other. In one respect, however, the stools used for the bacteriological analyses were peculiar. For this special work the first stool passed after 6 A. M. was employed, so that in general, where the individual passed more than one stool a day, the feces mployed for the bacteriological examination contained less water than the average. The error resulting from this cause would not seem to be very serious. It probably tends to render the values for the bacteria too high rather than too low.

Table 4, A to L, gives the data of these calculations. The daily number of fecal bacteria has been calculated in two ways; first, on the basis of the weight of the fresh feces, and second, on the basis of dry substance. Inasmuch as the number of bacteria per milligram varies with the dry substance in feces to a certain extent, the latter set of figures is the more accurate. Calculated on the basis of moist feces, the highest values, daily fecal bacteria by enumeration, occur in Subject I, July 3, 64×10^{12}, and Subject H, January 6, 63×10^{12}. In each of these instances the individual suffered from a rather marked diarrhea during the eight-day period concerned, serving to increase the amount of feces considerably above the average for that individual. For that reason these two estimates are unusual if not really abnormal. Subject H, April 25, shows a daily excretion of 60×10^{12} bacteria and on June 12, 58×10^{12} bacteria. The lowest values occur in Subject B, June 3, 16×10^{12} bacteria per day, Subject J, July 20, 17×10^{12} bacteria, and Subject B, April 16, 18×10^{12} bacteria. Of the 12 individuals, Subject H shows the highest average daily number of bacteria for the entire period of observation, namely 45×10^{12}. Subject L has an average of 41×10^{12}; Subject D, 40×10^{12}. The lowest value is

TABLE 4.

Daily Fecal Bacteria and Bacterial Nitrogen.

SUBJECT A.

LABORATORY No.	PERIOD	AVERAGE DAILY FECES Gm.	AVERAGE DAILY DRY SUBSTANCE Gm.	AVERAGE DAILY NITROGEN Gm.	DATE BACT. EXAM.	NUMBER OF BACTERIA				GRAVIMETRIC DETERMINATION			
						Calculated on Moist Feces		Calculated on Dry Feces		Dry Bacteria in Dry Substance Per cent	Daily Dry Bacteria Gm.	Daily Bacterial Nitrogen Gm.	Bacterial N. in Total Fecal N. Per cent
						Per Gram. $\times 10^9$	Daily $\times 10^9$	Per Gram. $\times 10^9$	Daily $\times 10^9$				
B110	Jan. 4–11	112.85	27.760	1.636	Jan. 6	244	27,535	.....	......	....	....	.....	....
B122	Jan. 20–27	106.45	25.268	1.605	Jan. 22	339	36,087	1,246	31,484	41.5	10.49	.....	....
B134	Feb. 5–12	89.48	25.915	1.479	Feb. 6	348	31,137	1,338	34,674	45.3	11.74	.....	....
B146	Feb. 21–28	70.06	20.028	1.141	Feb. 24	348	24,382	1,261	25,255	49.1	9.83	0.738	64.6
B158	Mar. 8–15	78.24	21.166	1.170	Mar. 11	447	34,972	1,987	42,057	28.1	5.95	0.446	38.1
B170	Mar. 24–31	52.65	16.656	0.915	Mar. 27	472	24,851	1,368	22,662	24.2	4.03	0.419	45.8
B182	Apr. 9–16	52.15	15.372	0.814	Apr. 10	407	21,225	1,394	21,429	19.0	2.92	0.348	42.8
B194	A. 25–M. 2	62.39	19.014	0.989	Apr. 27	476	29,697	1,345	25,574	26.3	5.00	0.540	54.6
B206	May 11–18	60.28	18.654	1.032	May 13	359	21,639	1,026	19,139	17.8	3.32	0.349	33.8
B218	M. 27–J. 3	70.08	19.250	1.081	May 28	460	32,235	1,386	26,681	35.3	6.80	0.673	62.2
B230	June 12–19	50.34	16.130	0.909	June 15	539	27,132	1,590	25,647	35.0	5.65	0.582	64.0
B242	J. 28–J. 5	96.21	23.135	1.408	July 1	472	45,413	1,360	31,464	25.9	5.99	0.611	33.2
B254	July 14–21	68.80	19.253	1.125	July 17	347	23,874	1,428	27,493	15.3	2.95	0.306	27.2
B266	July 22–29	62.33	19.550	1.172	July 31	441	27,485	1,339	26,177	19.6	3.83	0.437	37.3
Average..........		....	....	....	....	....	28,172	....	27,672	....	4.50*	0.474*	44.5*

SUBJECT B.

B114	Jan. 12–19	79.34	14.995	0.822	Jan. 12	248	33,651	.....	......	....	....	.....	....
B126	Jan. 20–27	94.99	19.367	0.969	Jan. 27	325	30,871	1,570	30,406	34.0	6.58	.....	....
B138	Feb. 13–20	112.72	18.923	1.111	Feb. 13	239	26,941	1,390	26,303	46.3	8.76	0.640	57.5
B150	F. 29–M. 7	97.14	15.598	0.966	Mar. 2	284	27,587	1,495	23,319	28.4	4.43	0.456	47.2
B162	Mar. 16–23	101.35	17.221	0.965	Mar. 16	296	30,000	1,794	30,894	26.6	4.58	0.481	49.9
B174	Apr. 1– 8	83.71	17.252	0.922	Apr. 1	366	30,639	1,634	28,190	....	....	....	....
B186	Apr. 9–16	43.97	9.533	0.509	Apr. 16	416	18,294	1,496	14,261	19.6	1.87	0.194	38.2
B198	A. 25–M. 2	77.44	15.104	0.843	May 1	379	29,349	1,458	22,022	20.0	3.02	0.329	39.1
B210	May 11–18	85.61	13.564	0.896	May 18	296	25,341	1,600	21,702	19.6	2.66	0.303	33.8
B222	M. 27–J. 3	53.61	10.359	0.540	June 3	298	15,977	1,935	20,045	27.1	2.81	0.334	61.9
B234	June 12–19	97.89	19.878	1.054	June 19	380	37,197	1,959	38,941	23.8	4.73	0.539	51.2

B246	July 6–13	103.20	19.096	1.009	July 6	425	43,860	1,683	32,139	27.8	5.31	0.589	58.4
B258	July 22–29	84.54	16.592	0.871	July 22	269	22,741	1,503	24,938	21.3	3.53	0.371	42.6
Average..........		....	....	....	....	....	28,650	....	26,097	....	3.56*	0.393*	46.9*
SUBJECT C.													
B104	D. 27–J. 3	80.86	21.216	1.304	Dec. 28	359	29,030	....	....	....	....	....	....
B116	Jan. 12–19	68.84	18.061	1.084	Jan. 14	405	27,879	....	....	....	....	....	....
B128	J. 28–F. 4	78.70	19.326	1.258	Jan. 29	402	31,637	....	....	....	....	....	....
B140	Feb. 13–20	Feces were not collected for this period			Feb. 20	457	....	2,642-	....	....	....	....	....
B154	F. 29–M. 7	76.48	16.587	1.159	Mar. 6	463	35,408	2,303	38,200	31.4	5.21	0.578	49.9
B166	Mar. 16–23	65.71	15.318	1.033	Mar. 23	436	28,651	1,434	21,966	38.8	5.94	0.672	65.0
B178	Apr. 1–8	68.65	16.880	1.100	Apr. 6	375	25,744	1,652	27,886	26.8	4.52	0.534	48.5
B190	Apr. 17–24	64.51	14.960	0.979	Apr. 22	461	29,739	1,897	28,379	26.5	3.96	0.444	45.3
B202	May 3–10	58.29	15.577	1.022	May 7	561	32,700	1,781	27,707	34.0	5.30	0.594	58.1
B214	May 19–26	63.80	15.484	1.048	May 23	457	29,157	1,554	24,062	28.0	4.34	0.490	46.8
B226	June 4–11	70.33	14.568	0.906	June 8	439	30,873	1,626	23,688	29.9	4.36	0.497	49.9
B238	June 20–27	84.48	17.281	1.189	June 25	678	57,274	2,166	37,430	28.6	4.94	0.568	47.8
B250	July 6–13	40.00	10.860	0.698	July 13	499	19,960	1,692	18,375	31.6	3.43	0.381	54.6
B262	July 22–29	77.77	19.028	1.257	July 27	388	30,175	1,665	31,682	28.4	5.40	0.594	47.3
Average..........		....	....	....	....	....	31,402	....	27,938	....	4.68*	0.530*	51.5*
SUBJECT D.													
B112	Jan. 4–11	165.40	31.241	1.767	Jan. 9	228	37,711	983	30,710	29.3	9.15	0.915	51.8
B123	Jan. 20–27	122.30	25.901	1.458	Jan. 25	262	32,042	992	25,694	45.9	11.89	1.189	....
B135	Feb. 5–12	147.78	27.793	1.713	Feb. 10	288	42,561	1,398	38,855	44.8	12.45	1.233	....
B147	Feb. 21–28	153.60	33.379	1.668	Feb. 27	273	41,933	1,125	37,551	25.9	8.65	0.900	53.9
B157	Mar. 8–15	133.36	30.102	1.582	Mar. 11	352	46,943	1,408	42,384	17.5	5.27	0.574	36.3
B169	Mar. 24–31	133.53	26.969	1.484	Mar. 27	322	42,997	1,594	42,989	24.3	6.55	0.786	53.0
B181	Apr. 9–16	128.14	25.236	1.461	Apr. 10	308	39,467	1,532	38,662	21.8	5.50	0.693	47.4
B193	A. 25–M. 2	110.93	26.158	1.420	Apr. 27	252	28,206	1,400	36,621	23.3	6.09	0.677	47.6
B205	May 11–18	120.90	25.113	1.534	May 13	328	39,655	1,624	40,784	24.0	6.03	0.639	41.6
B217	M. 27–J. 3	158.75	29.329	1.695	May 28	339	53,816	1,662	48,745	(34.9)	....	....	....
B229	June 12–19	117.85	28.883	1.563	June 15	265	31,230	1,417	40,927	25.3	7.31	0.818	52.3
B241	J. 28–J. 5	185.48	35.802	1.784	July 1	193	35,797	1,135	40,635	20.7	7.41	0.771	43.2
B253	July 14–21	122.75	27.108	1.398	July 17	307	37,684	1,471	39,876	13.3	3.61	0.411	29.4
B265	July 22–29	157.19	33.715	1.827	July 31	304	47.785	1,448	48,819	16.1	5.43	0.624	34.2
Average..........		....	....	....	....	....	39,845	....	39,518	....	5.99*	0.677*	43.6*

* Determinations previous to March 16 are not included in this average.

TABLE 4.—*Continued.*

SUBJECT E.

Laboratory No.	Period	Average Daily Feces Gm.	Average Daily Dry Substance Gm.	Average Daily Nitrogen Gm.	Date Bact. Exam.	Number of Bacteria				Gravimetric Determination			
						Calculated on Moist Feces		Calculated on Dry Feces		Dry Bacteria in Dry Substance Per cent	Daily Dry Bacteria Gm.	Daily Bacterial Nitrogen Gm.	Bacterial N. in Total Fecal N. Per cent
						Per Gram. $\times 10^9$	Daily $\times 10^9$	Per Gram. $\times 10^9$	Daily $\times 10^9$				
B118	Jan. 12–19	44.90	13.185	0.797	Jan. 16	454	20,385	....	.....	....	....	.....	....
B130	J 28–F. 4	82.00	23.106	1.378	Jan. 31	494	40,508	1,428	32,995	46.9	10.84	.....	....
B142	Feb. 13–20	99.19	23.325	1.477	Feb. 19	398	39,477	1,723	40,189	43.9	10.24	1.024	69.3
B149	F. 20–M. 7	81.44	23.354	1.442	Mar. 2	480	39,090	1,758	41,056	29.7	6.94	0.728	50.5
B161	Mar. 16–23	110.07	23.319	1.479	Mar. 16	298	32,802	1,574	36,704	14.5	3.38	0.345	23.3
B173	Apr. 1– 8	75.62	18.955	1.253	Apr. 1	535	40,459	2,011	38,119	....	....	....	....
B185	Apr. 9–16	110.85	23.937	1.621	Apr. 16	441	48,885	1,542	36,911	32.0	7.66	0.866	53.4
B197	A. 25–M. 2	67.11	15.053	1.056	May 5	514	31,412	1,586	23,874	24.0	3.61	0.383	36.3
B209	May 11–18	94.40	23.208	1.547	May 18	513	48,427	1,504	34 905	21.6	5.01	0.506	32.7
B221	M. 27–J. 3	78.05	19.117	1.283	June 3	343	26,771	1,478	28,255	31.7	6.06	0.691	53.8
B233	June 12–19	80.06	20.959	1.435	June 19	468	37,469	1,665	34,897	25.6	5.37	0.622	43.4
B245	July 6–13	84.47	17.995	1.202	July 6	373	31,507	1,581	28,450	30.4	5.47	0.624	51.9
B257	July 22–29	80.86	21.016	1.452	July 22	569	46,009	1,976	41,528	26.5	5.57	0.601	41.4
Average..........		....	....	....	....	....	37,352	....	34,824	....	5.27*	0.580*	42.0*

SUBJECT F.

B103	D. 27–J. 3	70.32	19.257	1.366	Dec. 28	459	32 277	....	....	....	....	....	....
B115	Jan. 12–19	66.34	19.730	1.396	Jan. 14	465	30,848		....	....	....	....	....
B127	J. 28–F. 4	52.94	15.815	1.069	Jan. 29	384	20,328	1,208	19,105	43.7	6.91	....	....
B139	Feb. 13–20	74.75	20.902	1.482	Feb. 17	469	30,368	1,209	25,271	39.8	8.32	0.765	51.6
B153	F. 29–M. 7	82.80	20.640	1.359	Mar. 6	263	21,776	1,229	25,367	34.4	7.10	0.753	55.4
B165	Mar. 16–23	66.79	17.163	1.172	Mar. 23	456	30,455	1,466	25,161	37.7	6.47	0.699	59.6
B177	Apr. 1–8	63.42	17.108	1.152	Apr. 6	465	29,493	1,838	31,445	28.2	4.82	0.574	49.8
B189	Apr. 17–24	78.14	18.846	1.296	Apr. 22	498	38,913	1,688	31,812	29.2	5.50	0.600	46.3
B201	May 3–10	70.12	18.490	1.216	May 7	476	33,380	1,566	28,955	40.0	7.40	0.770	63.3
B213	May 19–26	74.97	18.775	1.327	May 23	408	30,590	1,600	30,040	28.1	5.28	0.538	40.6
B225	June 4–11	66.50	17.670	1.238	June 8	477	31,720	1,647	30,212	25.2	4.46	0.454	36.7
B237	June 20–27	60.45	15.740	1.062	June 25	423	25,570	1,391	21,894	23.4	3.68	0.416	39.2

B249	July 6–13	73.52	17.687	1.209	July 13	418	30,733	1,679	29,696	27.8	4.92	0.556	46.0
B261	July 22–29	71.79	17.075	1.175	July 27	398	28,572	1,904	32,511	19.6	3.35	0.372	31.6
Average..........		....	....	....	....	....	29,645	....	27,622	....	5.10*	0.553*	45.9*
SUBJECT G.													
B107	D. 27–J. 3	97.84	25.885	1.439	Jan. 2	327	31,994	....	....	....	....	.....	....
B119	Jan. 20–27	104.11	25.866	1.090	Jan. 20	260	27,069	1,032	26,694	55.6	14.38	.....	....
B131	J. 28–F. 4	75.13	20.881	1.274	Feb. 3	379	28,472	1,243	25,955	51.8	....	.....	....
B143	Feb. 21–28	77.32	22.246	1.355	Feb. 21	534	41,289	1,786	39,731	43.4	9.65	0.946	69.6
B152	F. 29–M. 7	93.49	24.588	1.557	Mar. 4	480	44,875	1,579	38,824	37.0	9.10	0.900	57.8
B164	Mar. 16–23	80.75	21.391	1.384	Mar. 19	439	35,449	1,503	32,151	34.4	7.36	0.773	55.8
B176	Apr. 1– 8	69.95	20.102	1.291	Apr. 3	474	33,156	....	....	....	....	.....	....
B188	Apr. 17–24	90.20	23.931	1.550	Apr. 20	468	42,214	1,677	40,132	27.2	6.51	0.703	45.4
B200	May 3–10	98.30	25.090	1.710	May 4	446	43,842	1,522	38,187	33.0	8.28	0.886	51.8
B212	May 19–26	75.06	20.274	1.284	May 20	497	37,305	1,662	33,695	35.0	7.09	0.759	59.1
B224	June 4–11	91.98	21.872	1.453	June 5	290	26,674	1,812	39,632	22.8	4.99	0.584	40.2
B236	June 20–27	78.69	19.734	1.394	June 22	416	32,734	2,133	42,093	28.9	5.70	0.673	48.2
B248	July 6–13	67.89	19.593	1.191	July 9	483	32,791	1,548	30,330	28.1	5.51	0.584	49.0
B264	July 22–29	69.47	19.354	1.239	July 29	583	40,501	1,845	35,708	27.8	5.38	0.592	47.8
Average..........		....	....	....	....	....	36,387	....	35,261	....	6.35*	0.694*	49.7*
SUBJECT H.													
B109	Jan. 4–11	154.05	26.924	2.006	Jan. 6	412	63,469	2,157	58,075	37.7	10.15	.....	....
B121	Jan. 20–27	79.11	19.893	1.445	Jan. 22	383	30,299	1,157	23,016	48.0	9.55	.....	....
B133	Feb. 5–12	115.64	25.374	1.854	Feb. 6	364	42,093	1,583	40,167	44.9	11.39	.....	....
B145	Feb. 21–28	89.02	23.032	1.584	Feb. 24	451	40,150	1,646	37,911	....	....	.....	....
B156	Mar. 8–15	99.38	25.300	1.877	Mar. 10	518	51,479	1,853	46,881	37.5	9.49	1.063	56.6
B168	Mar. 24–31	82.91	19.882	1.474	Mar. 25	453	37,558	1,791	35,609	38.0	7.56	0.869	58.9
B180	Apr. 1– 8	88.11	23.045	1.660	Apr. 8	500	44,055	1,543	35,558	30.7	7.07	0.814	40.0
B192	A. 25–M. 2	92.35	22.411	1.769	Apr. 25	647	59,750	2,107	47,220	38.6	8.65	0.960	54.3
B204	May 11–18	84.50	21.128	1.573	May 11	456	38,532	1,466	30,974	30.7	6.49	0.694	44.1
B216	May 19–26	91.89	21.253	1.587	May 25	512	47,048	1,896	40,296	29.4	6.25	0.706	44.5
B228	June 12–19	82.59	22.758	1.677	June 12	707	58,391	2,230	50,750	40.2	9.15	1.006	60.0
B240	J. 28–J. 5	93.94	19.111	1.460	June 29	536	50,352	2,243	42,866	31.5	6.02	0.680	46.6
B252	July 14–21	97.40	20.508	1.495	July 15	322	31,363	1,905	39,068	29.9	6.13	0.717	48.0
B200	July 22–29	76.54	18.861	1.404	July 24	540	41,332	1,736	32,743	42.5	8.02	0.922	65.7
Average..........		....	....	....	....	....	45,419	....	40,081	....	7.26*	0.819*	52.3*

* Determinations previous to March 16 are not included in this average

TABLE 4.—*Continued.*

SUBJECT I.

LABORATORY No.	PERIOD	AVERAGE DAILY FECES	AVERAGE DAILY DRY SUBSTANCE	AVERAGE DAILY NITROGEN	DATE BACT. EXAM.	NUMBER OF BACTERIA				GRAVIMETRIC DETERMINATION			
						Calculated on Moist Feces		Calculated on Dry Feces		Dry Bacteria in Dry Substance	Daily Dry Bacteria	Daily Bacterial Nitrogen	Bacterial N. in Total Fecal N.
		Gm.	Gm.	Gm.		Per Gram. $\times 10^9$	Daily $\times 10^9$	Per Gram. $\times 10^9$	Daily $\times 10^9$	Per cent	Gm.	Gm.	Per cent
B113	Jan. 12–19	67.72	18.338	1.063	Jan. 12	335	22,686	....	......	....	....	.....	....
B125	Jan. 20–27	95.14	22.641	1.310	Jan. 27	290	27,591	1,401	31,720	13.3	3.01	.....	....
B137	Feb. 13–20	96.18	22.664	1.439	Feb. 14	498	47,898	1,772	40,161	44.1	9.99	0.919	63.9
B160	Mar. 8–15	Feces	were not collected	for this period	Mar. 14	282	......	1,476	......	24.0	....	.....	....
B172	Mar. 24–31	47.11	10.298	0.638	Mar. 31	442	20,823	1,693	17,435	....	....	.....	....
B184	Apr. 9–16	77.90	20.881	1.260	Apr. 13	399	31,082	1,541	32,178	35.7	7.45	0.842	66.8
B196	A. 25–M. 2	58.08	16.424	0.986	Apr. 29	457	26,543	1,465	24,061	24.0	3.94	0.429	43.6
B208	May 11–18	53.45	14.551	0.884	May 15	478	25,549	1,935	28,156	37.3	5.43	0.554	62.6
B220	M. 27–J. 3	82.04	21.570	1.324	June 1	397	32,570	1,582	34,124	35.8	7.72	0.831	62.7
B232	June 12–19	67.67	14.545	0.860	June 17	492	33,296	1,751	25,468	....	....	.....	....
B244	J. 28–J. 5	104.76	24.129	1.245	July 3	612	64,113	2,250	54,290	28.8	6.95	0.744	59.6
B256	July 14–21	88.40	18.115	1.246	July 20	576	50,918	2,133	38,639	30.8	5.58	0.642	51.5
Average..........		....	....	....	....	....	36,038	....	32,646	....	6.18*	0.674*	57.8*

SUBJECT J.

LABORATORY No.	PERIOD	AVERAGE DAILY FECES	AVERAGE DAILY DRY SUBSTANCE	AVERAGE DAILY NITROGEN	DATE BACT. EXAM.	Moist Feces Per Gram. $\times 10^9$	Moist Feces Daily $\times 10^9$	Dry Feces Per Gram. $\times 10^9$	Dry Feces Daily $\times 10^9$	Dry Bacteria in Dry Substance	Daily Dry Bacteria	Daily Bacterial Nitrogen	Bacterial N. in Total Fecal N.
B111	Jan. 4–11	136.54	23.725	1.447	Jan. 9	298	40,689	1,096	26,003	28.5	6.76	0.676	46.7
B124	Jan. 20–27	69.51	18.169	1.098	Jan. 25	309	21,479	1,128	20,495	48.1	8.74	0.874	79.6
B136	Feb. 5–12	137.98	23.817	1.563	Feb. 10	220	30,356	1,392	33,153	44.6	10.62	1.062	68.0
B148	Feb. 21–28	98.90	20.915	1.356	Feb. 27	386	38,175	1,317	27,545	45.5	9.52	0.952	70.2
B159	Mar. 8–15	89.83	19.049	1.167	Mar. 13	364	32,698	1,291	24,592	28.1	5.35	0.535	45.9
B171	Mar. 24–31	62.86	15.917	0.967	Mar. 30	401	25,207	1,373	21,854	34.4	5.48	0.559	57.8
B183	Apr. 9–16	66.20	15.694	0.990	Apr. 14	439	29,062	1,287	20,198	26.8	4.21	0.450	45.5
B195	A. 25–M. 2	92.63	18.943	1.213	Apr. 29	302	27,974	1,302	24,664	35.0	6.63	0.709	58.5
B207	May 11–18	88.38	18.084	1.206	May 15	383	33,850	1,710	30,924	38.8	7.02	0.730	60.5
B219	M. 27–J. 3	70.64	17.423	1.079	June 1	476	33,625	1,817	31,658	34.5	6.01	0.619	57.4
B231	June 12–19	76.14	17.840	1.118	June 17	368	28,020	1,484	26,475	....	....	.....	....
B243	J. 28–J. 5	102.04	17.768	1.222	July 4	372	37,959	1,738	30,881	23.4	4.16	0.437	35.7
B255	July 14–21	76.74	16.158	1.068	July 20	228	17,497	1,416	22,880	19.2	3.10	0.335	31.3
Average..........		....	....	....	....	....	30,507	....	26,256	....	5.23*	0.548*	49.5*

SUBJECT K.

B105	D. 27–J. 3	195.25	27.450	1.802	Dec. 30	227	44,322	....	......	....	....	.....	....
B117	Jan. 12–19	135.40	24.204	1.592	Jan. 16	264	35,746	....	......	....	....	.....	....
B129	Jan. 28–31	175.88	26.312	1.654	Jan. 31	300	52,764	1,327	34,916	40.0	....	.....	....
B141	Feb. 13–20	156.94	25.374	1.527	Feb. 19	198	31,074	1,131	28,698	36.7	9.31	0.875	57.3
B151	F. 29–M. 7	123.27	21.890	1.387	Mar. 4	385	47,461	1,504	32,923	23.2	5.08	0.569	41.0
B163	Mar. 16–23	117.92	21.283	1.374	Mar. 19	358	42,217	1,914	40,736	19.0	4.04	0.420	30.6
B175	Apr. 1– 8	102.61	19.746	1.210	Apr. 3	282	28,937	....	......	....	....	....	....
B187	Apr. 17–24	134.95	21.593	1.445	Apr. 20	356	48,042	1,465	31,634	28.4	6.13	0.644	44.5
B199	May 3–10	118.12	20.099	1.252	May 4	260	30,713	1,244	25,003	17.4	3.50	0.364	29.1
B211	May 19–26	146.40	24.100	1.531	May 20	276	40,406	1,401	33,764	15.3	3.69	0.413	27.0
B223	June 4–11	129.26	22.107	1.580	June 5	317	40,976	1,294	28,606	15.4	3.40	0.368	23.3
B235	June 20–27	130.75	20.271	1.400	June 23	371	48,508	1,492	30,244	21.7	4.40	0.440	31.4
B247	July 6–13	128.46	22.157	1.434	July 9	224	28,775	1,514	33,546	22.5	4.99	0.509	35.5
B263	July 22–29	124.34	21.470	1.451	July 29	252	31,334	1,565	33,601	20.8	4.47	0.518	35.7
Average..........		....	....	....	....	....	37,492	....	32,152	....	4.33*	0.460*	32.1*

SUBJECT L.

B108	D. 27–J. 3	144.31	25.984	1.700	Jan. 2	331	47,768	....	.....	....	....	.....	....
B120	Jan. 20–27	153.92	23.566	1.924	Jan. 20	175	26,937	1.101	25,946	39.6	9.33	.....	....
B132	J. 28–F. 4	111.89	25.463	1.673	Feb. 3	203	22,713	1,269	32,313	43.3	11.03	.....	....
B144	Feb. 21–28	99.70	22.324	1.494	Feb. 21	370	36,889	1,331	29,713	43.2	9.64	0.945	63.3
B155	Mar. 8–15	99.30	20.965	1.462	Mar. 9	479	47,565	1,241	26,018	28.6	6.00	0.678	46.3
B167	Mar. 24–31	108.50	21.459	1.446	Mar. 25	340	36,890	1,744	37,424	23.1	4.96	0.506	35.0
B179	Apr. 1– 8	124.27	23.314	1.591	Apr. 8	390	48,467	1,674	39,028	28.4	6.62	0.775	48.7
B191	Apr. 17–24	132.84	23.482	1.606	Apr. 24	353	46,892	1,689	39,661	19.8	4.65	0.530	33.0
B203	May 11–18	107.04	20.866	1.477	May 11	367	39,283	1,631	34,032	24.2	5.05	0.586	39.7
B215	May 19–26	134.15	25.208	1.763	May 25	302	40,513	1,659	41,820	14.0	3.53	0.416	23.6
B227	June 12–19	110.21	23.288	1.633	June 12	446	49,154	2,134	49,697	36.1	8.41	0.925	56.6
B239	J. 28–J. 5	125.77	23.495	1.713	June 29	341	42,888	1,655	38,884	27.3	6.41	0.712	41.6
B251	July 14–21	112.07	23.296	1.646	July 15	430	48,190	1,928	44,915	30.9	7.20	0.785	47.7
B259	July 22–29	79.72	18.159	1.233	July 25	507	40,418	1,980	35,955	21.5	3.90	0.418	33.9
Average..........		....	....	....	....	....	41,041	....	36,570	....	5.64*	0.628*	40.0*

* Determinations previous to March 16 are not included in this average.

in Subject A, 28×10^{12}, and in Subject B, 29×10^{12}. The average number of daily fecal bacteria for the 12 individuals is 35×10^{12}

In those instances where the average daily number of bacteria was also calculated upon the basis of fecal dry substance, the values are in general slightly lower than those calculated upon the basis of moist feces, and the range of variation is not quite so wide. The highest values for daily fecal bacteria, calculated on the basis of dry substance, occur in Subject H, January 6, 58×10^{12} bacteria per day, and Subject I, July 3, 54×10^{12}. A value of 51×10^{12} occurs in Subject H on June 12. The lowest values occur in Subject B, April 16, 14×10^{12} bacteria per day, and in Subject I, March 31, 17×10^{12} bacteria per day. Of the 12 individuals, Subject H still shows the highest average value, 40×10^{12}; Subject B shows the lowest, 26×10^{12} bacteria per day. The grand average of the 12 subjects for all examinations in which the number of daily bacteria was calculated on the basis of dry substance, is 32×10^{12} bacteria per day, about 9 per cent lower than when calculated on the basis of moist feces.

The daily bacterial dry substance of the feces has been calculated from the Strasburger determinations on the basis of dry substance throughout. The highest daily bacterial dry substance is shown by Subject H, June 12, 9.15 gm., containing 1.006 gm. of nitrogen. On April 25, the same individual excreted 8.65 gm. of dry bacterial substance, containing 0.960 gm. of nitrogen. The lowest value is in Subject B, on April 16, 1.87 gm. of dry bacterial substance containing 0.194 gm. of nitrogen. This same subject excreted on May 18, 2.66 gm., and on June 3, 2.81 gm. of dry bacterial substance containing 0.303 and 0.334 gm. of nitrogen respectively. For the whole period of observation, Subject H shows the highest average daily bacterial dry substance, 7.26 gm. containing 0.819 gm. of nitrogen; Subject B shows the lowest average, 3.56 gm. dry bacterial substance containing 0.393 gm. of nitrogen. The average of the 12 subjects is 5.34 gm. daily bacterial dry substance, representing 0.585 gm. of nitrogen.

The portion of the total fecal nitrogen contained in the fecal bacteria varied in the single examinations from 66.8 per cent in Subject I, April 13, to 23.3 per cent in Subject K, June 5. Subject I shows the highest average value for all examinations, 57.4 per cent, Subject K the lowest, 32.1 per cent. The grand average for the 12 individuals is 46.3 per cent of the total fecal nitrogen, contained in the fecal bacteria.

The following table, 5, summarizes briefly the extreme variations and the mean of these observations.

TABLE 5.

SUMMARY OF DAILY FECAL BACTERIA.

	Number Calculated on		Dry Bacterial Substance	Bacterial Nitrogen	
	Moist Feces	Dry Feces	Grams	Daily Quantity Grams	In Total Fecal Nitrogen Per cent
Mean	35×10^{12}	32×10^{12}	5.34	0.585	46.3
Maximum	64×10^{12}	58×10^{12}	9.15	1.006	66.8
Minimum	16×10^{12}	14×10^{12}	1.87	0.194	23.3

DIFFERENTIAL COUNTS.

Strasburger[20] has studied Gram stained microscopic preparations of adult feces. He found the bacteria predominantly Gram negative, especially when the diet consisted largely of milk and vegetables. In the case of a meat diet he found an increased number of Gram positive micrococci.

Table 6, A to L, shows the results of differential counts of Gram stained preparations of mixed fecal flora from 157 different stools. The classes of bacteria, designated in the table, were in general quite easily distinguished from each other except where the distinction rested solely upon the color. In these cases there was some difficulty to which attention has been directed in describing the method of staining.

Bacilli 0.4 to 0.9 μ wide and from 1.0 to 2.5 μ long were classed as *B. coli* type. The quantity of these varied from 81.6 per cent in Subject L, January 20, 1908, to 19 per cent in Subject E, April 1, 1908. In the second class, all the remaining Gram negative rods were placed. The quantity of these varied from 46.2 per cent in Subject E on March 2, 1908, to 0.4 per cent in Subject H on January 6, 1908. The distinction between these two classes is not sharp. The Gram negative micrococci varied from 36.8 per cent in Subject H on January 6, to 0.8 per cent in Subject L on June 12, 1908. The distinction between the bacteria of this class and the bacteria classified as Gram positive cocci was not accurate and frequently exceedingly difficult. Spirilla

TABLE 6.

Differential Count of Fecal Bacteria.

SUBJECT A.

Laboratory No......	B110	B122	B134	B146	B158	B170	B182	B194	B206	B218	B230	B242	B254	B266	Average
Date..............	Jan. 6	Jan. 22	Feb. 6	Feb. 24	Mar. 11	Mar. 27	Apr. 10	Apr. 27	May 13	May 28	June 15	July 1	July 17	July 31	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type....	47.8	54.0	62.6	36.0	42.6	38.7	39.8	32.8	45.4	33.4	36.6	43.8	37.2	Count not made Poor stain	42.4
Other negative rods.	12.3	14.6	17.6	36.2	22.6	36.5	32.8	37.8	26.8	31.6	32.8	24.4	27.8		27.2
Negative cocci......	12.8	7.5	9.0	4.4	5.2	1.4	1.6	5.8	2.4	4.4	7.8	6.0	3.8		5.6
Negative spirochetes.	0.0	0.0	0.4	0.4	0.8	0.4	0.0	0.0	0.6	0.0	0.0	0.0	3.2		0.4
Gram positive rods..	8.1	11.9	3.8	5.6	6.2*	5.0	8.8*	8.0	4.2	4.6	3.8	6.8	3.8		6.2
Positive cocci.......	18.8	9.2	0.4	3.8	15.4	14.2	10.4	10.8	10.2	16.8	15.6	16.4	19.4		12.4
Oval free spores.....	0.0	1.2	4.8	5.0	3.4	3.5	4.8	3.8	8.2	8.6	3.2	2.4	4.0		4.1
Spherical free spores.	0.2	1.6	1.4	8.6	3.8	0.3	1.8	1.0	2.2	0.6	0.2	0.2	0.8		1.7
Total Gram negative	72.9	76.1	89.6	77.0	71.2	77.0	74.2	76.4	75.2	69.4	77.2	74.2	72.0		75.6
Total Gram positive.	26.9	21.1	4.2	9.4	21.6	19.2	19.2	18.8	14.4	21.4	19.4	23.2	23.2		18.6
Total free spores....	0.2	2.8	6.2	13.6	7.2	3.8	6.6	4.8	10.4	9.2	3.4	2.6	4.8		5.8
Total negative rods..	60.1	68.6	80.2	72.2	65.2	75.2	72.6	70.6	72.2	65.0	69.4	68.2	65.0		69.6
Total micrococci....	31.6	16.7	9.4	8.2	20.6	15.6	12.0	16.6	12.6	21.2	23.4	22.4	23.2		18.0
Quality of the stain..	....	....	....	Poor	....	Fair	Poor	Exc.	Good	Good	Good	Good	Fair		....

SUBJECT B.

Laboratory No......	....	B114	B126	B138	B150	B162	B174	B186	B198	B210	B222	B234	B246	B258	Average
Date..............	....	Jan. 12	Jan. 27	Feb. 13	Mar. 2	Mar. 16	Apr. 1	Apr. 16	May 1	May 18	June 3	June 19	July 6	July 22	
		Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type....	....	44.0	39.4	37.2	31.6	42.2	40.4	41.2	39.6	44.4	39.4	31.6	35.4	41.6	39.1
Other negative rods.	....	29.0	21.4	17.6	31.8	26.4	25.8	24.8	32.6	34.4	28.0	34.6	36.6	30.2	28.7
Negative cocci......	....	18.4	26.2	19.2	11.0	21.8	9.8	6.6	5.8	4.8	3.6	2.2	5.6	4.8	10.7
Negative spirochetes.	....	0.0	1.4	3.8	2.2	0.6	0.2	0.4	0.0	0.4	0.4	0.0	0.0	0.6	0.8
Gram positive rods .	....	4.8	5.4*	4.6	5.0	3.4	2.4	3.4	4.4	6.0	7.2	4.0	6.8	5.2	4.8
Positive cocci.......	....	3.2	6.0	16.8	17.8	4.0	20.8	22.6	17.0	9.4	20.8	26.8	15.0	17.0	15.2
Oval free spores....	....	0.6	0.2	0.6	0.2	0.4	0.0	0.8	0.0	0.6	0.4	0.4	0.4	0.6	0.4
Spherical free spores.	....	0.0	0.0	0.2	0.4	1.2	0.6	0.2	0.6	0.0	0.2	0.4	0.2	0.6	0.3
Total Gram negative	....	91.4	88.4	77.8	76.6	91.0	76.2	73.0	78.0	84.0	71.4	68.4	77.6	77.2	79.3
Total Gram positive.	....	8.0	11.4	21.4	22.8	7.4	23.2	26.0	21.4	15.4	28.0	30.8	21.8	22.2	20.0
Total free spores....	....	0.6	0.2	0.8	0.6	1.6	0.6	1.0	0.6	0.6	0.6	0.8	0.6	0.6	0.7
Total negative rods.	....	73.0	60.8	54.8	63.4	68.6	66.2	66.0	72.2	78.8	67.4	66.2	72.0	71.8	67.8
Total micrococci....	....	21.6	32.2	36.0	28.8	25.8	30.6	29.2	22.8	14.2	24.4	29.0	20.6	21.8	25.9
Quality of the stain.	....	....	....	....	Good	....	Good	Fair	Poor	Good	Good	Good	Good	Good	

SUBJECT C.

Laboratory No.	B104	B116	B128	B140	B154	B166	B178	B190	B202	B214	B226	B238	B250	B262	Average
Date	Dec. 28	Jan. 14	Jan. 29	Feb 20	Mar. 6	Mar. 23	Apr. 6	Apr. 22	May 7	May 23	June 8	June 25	July 13	July 27	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type	62.6	67.0	55.2	51.2	53.6	52.0	35.6	54.0	33.4	39.4	39.8	55.4	39.0	36.8	48.2
Other negative rods	20.0	8.2	11.8	27.0	26.4	35.0	30.2	31.2	33.3	26.0	32.0	16.0	35.2	25.6	25.6
Negative cocci	7.2	10.8	2.0	7.5	4.8	1.8	5.6	5.6	7.4	6.0	5.4	3.2	4.2	1.2	5.2
Negative spirochetes	1.2	2.4	0.2	0.0	5.2	4.6	4.4	1.2	1.7	0.4	1.0	1.0	0.4	1.4	1.8
Gram positive rods	2.6	3.4	11.4	5.0	2.2	2.6	5.0	2.4	4.7	6.8	5.0	5.2	3.4	2.4	4.4
Positive cocci	4.6	2.6	18.8	8.8	6.8	3.2	17.6	2.8	18.0	19.6	15.2	17.0	16.8	31.6	13.1
Oval free spores	1.0	3.4	0.4	0.3	1.0	0.8	1.0	2.8	1.2	1.4	1.4	1.6	1.0	0.4	1.3
Spherical free spores	0.8	2.2	0.2	0.2	0.0	0.0	0.6	0.0	0.3	0.2	0.2	0.6	0.0	0.6	0.4
Total Gram negative	91.0	88.4	69.2	85.7	90.0	93.4	75.8	92.0	75.8	72.0†	78.2	75.6	78.8	65.0	80.8
Total Gram positive	7.2	6.0	30.2	13.8	9.0	5.8	22.6	5.2	22.7	26.4	20.2	22.2	20.2	34.0	17.5
Total free spores	1.8	5.6	0.6	0.5	1.0	0.8	1.6	2.8	1.5	1.6	1.6	2.2	1.0	1.0	1.7
Total negative rods	82.6	75.2	67.0	78.2	80.0	87.0	65.8	85.2	66.7	65.4	71.8	71.4	74.2	62.4	73.8
Total micrococci	11.8	13.4	20.8	16.3	11.6	5.0	23.2	8.4	25.4	25.6	20.6	20.2	21.0	32.8	18.3
Quality ofthe stain	....	....	....	....	Poor	Good	Fair	Poor	Good	Good	Exc.	Fair	Good	Fair	

SUBJECT D.

Laboratory No.	B112	B123	B135	B147	B157	B169	B181	B193	B205	B217	B229	B241	B253	B265	Average
Date	Jan. 9	Jan. 25	Feb. 10	Feb. 27	Mar. 11	Mar. 27	Apr. 10	Apr. 27	May 13	May 28	June 15	July 1	July 17	July 31	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type	67.6	49.2	48.4	40.0	45.8	44.0	42.8	53.8	46.4	47.4	34.6	50.6	42.2	28.4	45.8
Other negative rods	2.0	16.0	18.0	27.8	20.6	27.0	20.4	20.8	20.6	29.6	34.4	23.6	35.2	40.0	24.0
Negative cocci	12.8	16.0	22.4	14.6	17.4	10.8	8.8	6.6	6.8	3.6	3.0	4.2	5.0	6.2	9.9
Negative spirochetes	0 0	0.4	1.6	1.4	1.0	2.0	1.6	2.0	7.6	0.0	3.6	5.8	0.0	7.2	2.4
Gram positive rods	8.2	7.8	4.4	11.0	6.4	4.0	8.4	8.0	9.0	8.0	9.2	7.6	7.6	3.6	7.4
Positive cocci	8.6	9.4	3.4	5.0	6.6	11.0	18.0	8.4	9.2	10.6	13.4	8.0	9.2	13.6	9.6
Oval free spores	0.6	0.4	0.4	0.2	0.2	0.0	0.0	0.4	0.4	0.6	0.6	0.0	0.4	0.8	0.4
Spherical free spores	0.2	0.8	1.4	0.0	2.0	1.2	0.0	0.0	0.0	0.2	1.2	0.2	0.4	0.2	0.5
Total Gram negative	82.4	81.6	90.4	83.8	84.8	83.8	73.6	83.2	81.4	80.6	75.6	84.2	82.4	81.8	82.1
Total Gram positive	16.8	17.2	7.8	16.0	13.0	15.0	26.4	16.4	18.2	18.6	22.6	15.6	16.8	17.2	17.0
Total free spores	0.8	1.2	1.8	0.2	2.2	1.2	0.0	0.4	0.4	0.8	1.8	0.2	0.8	1.0	0.9
Total negative rods	69.6	65.2	66.4	67.8	66.4	71.0	63.2	74.6	67.0	77.0	69.0	74.2	77.4	68.4	69.8
Total microccocci	21.4	25.4	25.8	19.6	24.0	21.8	26.8	15.0	16.0	14.2	16.4	12.2	14.2	19.8	19.5
Quality of the stain	....	....	....	Good	Good	Good	Good	Good	Good	Good	Good	Fair	Fair	Good	

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* One of these bacilli (0.2 per cent. of the bacteria counted) contained a spore.

† A single Gram negative spirillum was observed in counting 500 bacteria in this stool. This 0.2 per cent. is included in the total Gram negative bacteria.

TABLE 6 (*Continued*).

SUBJECT E.

Laboratory No......	B106	B118	B130	B142	B149	B161	B173	B185	B197	B209	B221	B233	B245	B257	Average
Date..............	Dec. 30	Jan. 16	Jan. 31	Feb. 19	Mar. 2	Mar. 16	Apr. 1	Apr. 16	May 1	May 18	June 3	June 19	July 6	July 22	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type....	54.0	63.2	Count not made	42.8	29.6	42.8	19.0	48.4	53.0	Count not made	39.2	39.8	60.2	31.4	43.6
Other negative rods..	16.6	8.2		38.4	46.2	31.6	45.0	28.6	31.4		30.2	33.8	27.8	38.2	31.3
Negative cocci......	10.0	13.2		12.2	8.4	5.6	6.0	8.8	6.6		6.4	3.8	5.2	5.4	7.6
Negative spirochetes.	2.0	0.8		0.2	0.2	0.0	0.0	0.2	0.6		0.0	0.0	0.6	0.8	0.5
Gram positive rods..	6.6	2.4		1.6	2.8*	0.0	4.2	3.8	2.4		7.8	3.0	2.0	3.8	3.8
Positive cocci.......	9.6	7.2		1.2	12.0	13.0	24.2	7.8	4.6		14.2	18.2	3.0	18.4	11.2
Oval free spores.....	0.6	2.2		2.0	0.6	0.6	1.0	2.2	0.6		1.8	1.2	0.4	2.0	1.3
Spherical free spores.	0.6	2.8		1.6	0.2	0.4	0.6	0.2	0.8		0.4	0.2	0.8	0.0	0.7
Total Gram negative	82.6	85.4		93.6	84.4	80.0	70.0	86.0	91.6		75.8	77.4	93.8	75.8	83.0
Total Gram positive.	16.2	9.6		2.8	14.8	19.0	28.4	11.6	7.0		22.0	21.2	5.0	22.2	15.0
Total free spores....	1.2	5.0		3.6	0.8	1.0	1.6	2.4	1.4		2.2	1.4	1.2	2.0	2.0
Total negative rods..	70.6	71.4		81.2	75.8	74.4	64.0	77.0	84.4		69.4	73.6	88.0	69.6	74.9
Total micrococci....	19.6	20.4		13.4	20.4	18.6	30.2	16.6	11.2		20.6	22.0	8.2	23.8	18.8
Quality of the stain..	....	....		Good	Good	....	Good	Poor	Poor		Good	Good	Fair	Good	

SUBJECT F.

Laboratory No......	B103	B115	B127	B139	B153	B165	B177	B189	B201	B213	B225	B237	B249	B261	Average
Date..............	Dec. 28	Jan. 14	Jan. 29	Feb. 17	Mar. 6	Mar. 23	Apr. 6	Apr. 22	May 7	May 23	June 8	June 25	July 13	July 27	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type....	53.4	44.8	41.4	Count not made	40.6	41.6	52.4	42.4	42.8	39.4	45.2	37.8	46.0	44.4	44.0
Other negative rods..	22.6	23.0	14.8		14.6	20.2	18.0	25.8	20.0	30.6	21.8	27.8	32.4	25.4	22.9
Negative cocci......	10.4	14.8	20.6		21.4	17.2	10.0	10.8	5.2	3.8	7.2	6.4	3.8	7.4	10.7
Negative spirochetes.	1.0	0.0	1.0		0.4	0.8	0.0	0.6	0.6	0.0	0.0	0.0	0.0	0.0	0.3
Gram positive rods..	2.2	7.4	10.4		11.0	8.0	6.2	10.0	12.0	9.2	8.0	6.2	6.4	11.6	8.4
Positive cocci.......	8.0	7.8	9.6		10.6	10.2	11.2	8.8	17.0	15.8	17.0	19.2	11.0	10.6	12.0
Oval free spores.....	1.6	1.6	0.8		0.4	0.6	1.8	0.4	1.0	0.4	0.8	1.4	0.4	0.2	0.9
Spherical free spores.	0.8	0.6	1.4		1.0	1.4	0.4	1.2	1.4	0.8	0.0	1.2	0.0	0.4	0.8
Total Gram negative	87.4	82.6	77.8		77.0	79.8	80.4	79.6	68.6	73.8	74.2	72.0	82.2	77.2	77.9
Total Gram positive.	10.2	15.2	20.0		21.6	18.2	17.4	18.8	29.0	25.0	25.0	25.4	17.4	22.2	20.4
Total free spores....	2.4	2.2	2.2		1.4	2.0	2.2	1.6	2.4	1.2	0.8	2.6	0.4	0.6	1.7
Total negative rods .	76.0	67.8	56.2		55.2	61.8	70.4	68.2	62.8	70.0	67.0	65.6	78.4	69.8	66.9
Total micrococci....	18.4	22.6	30.2		32.0	27.4	21.2	19.6	22.2	19.6	24.2	25.6	14.8	18.0	22.7
Quality of the stain..	....	....	....		....	Fair	Poor	Good	Good	Good	Good	Good	Fair	Good	

SUBJECT G.

Laboratory No......	....	B119	B131	B143	B152	B164	B176	B188	B200	B212	B224	B236	B248	B264	Average
Date.............	....	Jan. 20	Feb. 3	Feb. 21	Mar. 4	Mar. 19	Apr. 3	Apr. 20	May 4	May 20	June 5	June 22	July 9	July 29	
		Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type....	....	46.0	47.6	51.8	30.0	32.4	34.6	39.4	63.2	52.4	52.0	42.0	41.2	54.2	45.1
Other negative rods.	....	16.0	22.4	17.6	29.2	30.8	32.0	19.8	22.0	20.8	25.2	28.8	32.3	27.8	25.0
Negative cocci......	....	11.8	10.8	5.6	3.2	7.4	8.4	5.2	4.4	9.4	5.6	3.8	11.2	10.4	7.5
Negative spirochetes	....	3.2	3.4	1.0	1.0	1.6	0.6	1.6	0.2	0.2	0.2	0.6	0.3	0.6	1.1
Gram positive rods.	....	6.4†	4.4†	5.8	11.6‡	7.6	4.4	5.8‡	2.0	4.8	5.0	5.0	5.2	1.0	5.3§
Positive cocci.......	....	10.6	4.4	13.0	21.6	16.8	15.6	26.0	6.2	9.4	8.6	17.4	8.1	4.0	12.4
Oval free spores....	....	1.6	4.4	2.6	1.4	2.6	4.4	2.2	1.0	2.8	3.0	2.0	1.5	1.6	2.4
Spherical free spores	....	4.4	2.6	2.6	2.0	0.8	0.0	0.0	1.0	0.2	0.4	0.4	0.2	0.4	1.2
Total Gram negative	....	77.0	84.2	76.0	63.4	72.2	75.6	66.0	89.8	82.8	83.0	75.2	85.0	93.0	78.7
Total Gram positive.	....	17.0	8.8	18.8	33.2	24.4	20.0	31.8	8.2	14.2	13.6	22.4	13.3	5.0	17.7
Total free spores....	....	6.0	7.0	5.2	3.4	3.4	4.4	2.2	2.0	3.0	3.4	2.4	1.7	2.0	3.6
Total negative rods.	....	62.0	70.0	69.4	59.2	63.2	66.6	59.2	85.2	73.2	77.2	70.8	73.5	82.0	70.1
Total micrococci....	....	22.4	15.2	18.6	24.8	24.2	24.0	31.2	10.6	18.8	14.2	21.2	19.3	14.4	19.9
Quality of the stain.	....	....	Fair	....	Good	Good	Fair	Good	Fair	Good	Fair	Exc.	Good	Poor	

SUBJECT H.

Laboratory No......	B109	B121	B133	B145	B156	B168	B180	B192	B204	B216	B228	B240	B252	B260	Average
Date..............	Jan. 6	Jan. 22	Feb. 6	Feb. 24	Mar. 10	Mar. 25	Apr. 8	Apr. 25	May 11	May 25	June 12	June 29	July 15	July 24	
	Per cent	P er cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type....	58.4	44.6	48.4	43.8	43.4	51.6	Count not made	48.2	45.6	47.2	32.8	48.4	50.0	48.8	47.0
Other negative rods.	0.4	12.2	16.4	18.6	35.4	33.2		28.0	29.6	31.2	34.2	23.4	24.2	27.6	24.2
Negative cocci......	36.8	11.2	22.8	18.8	3.6	4.8		7.0	3.6	4.8	12.0	4.4	4.4	12.2	11.2
Negative spirochetes.	0.0	1.8	0.4	1.2	1.8	0.4		0.6	0.8	0.2	0.0	0.0	0.0	0.4	0.6
Gram positive rods..	0.0	13.0	6.2	7.0	4.2	2.8		3.2	5.2	5.6	5.6	4.0	3.8	1.8	4.8
Positive cocci.......	2.6	15.8	5.6	9.4	9.2	5.6		11.4	13.0	7.8	14.0	18.4	16.0	7.8	10.5
Oval free spores.....	1.4	0.8	0.0	0.4	2.0	1.4		1.2	2.0	3.0	1.4	1.4	1.4	1.4	1.4
Spherical free spores.	0.4	0.6	0.2	0.8	0.4	0.2		0.4	0.2	0.2	0.0	0.0	0.2	0.0	0.3
Total Gram negative	95.6	69.8	88.0	82.4	84.2	90.0		83.8	79.6	83.4	79.0	76.2	78.6	89.0	83.0
Total Gram positive.	2.6	28.8	11.8	16.4	13.4	8.4		14.6	18.2	13.4	19.6	22.4	19.8	9.6	15.3
Total free spores....	1.8	1.4	0.2	1.2	2.4	1.6		1.6	2.2	3.2	1.4	1.4	1.6	1.4	1.7
Total negative rods..	58.8	56.8	64.8	62.4	78.8	84.8		76.2	75.2	78.4	67.0	71.8	74.2	76.4	70.1
Total micrococci....	39.4	27.0	28.4	28.2	12.8	10.4		18.4	16.6	12.6	26.0	22.8	20.4	20.0	21.7
Quality of the stain..	....	....	Good	Fair	....	Good		Fair	Good	Fair	Good	Good	Fair	Fair	

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* Four of these bacilli (0.8 per cent. of the bacteria counted) contained spores.

† Three of these bacilli (0.6 per cent. of the bacteria counted) contained spores.

‡ Two of these bacilli (0.4 per cent. of the bacteria counted) contained spores.

§ One of these bacilli (0.2 per cent. of the bacteria counted) contained a spore.

TABLE 6 (*Continued*).

SUBJECT I

Laboratory No.	....	B101	B113	B125	B137	B160	B172	B184	B196	B208	B220	B232	B244	B256	Average
Date	....	Dec. 26	Jan. 12	Jan. 27	Feb. 14	Mar. 14	Mar. 31	Apr. 13	Apr. 29	May 15	June 1	June 17	July 3	July 20	
		Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacilus coli type	....	63.0	69.0	69.8	48.4	50.4	46.2	46.2	38.8	43.0	42.4	45.0	34.0	43.0	49.2
Other negative rods	....	14.6	3.6	16.4	35.8	14.8	19.4	24.6	34.6	27.0	30.8	30.0	44.4	34.2	25.4
Negative cocci	....	10.2	9.8	2.6	2.4	19.0	9.4	7.2	6.0	6.0	5.2	1.4	6.0	4.4	6.9
Negative spirochetes	....	0.2	1.4	0.0	0.8	0.6	0.2	0.4	0.2	0.8	0.0	0.0	0.0	0.0	0.4
Gram positive rods	....	2.9	1.6	2.8	3.2	7.6	4.8	6.2	8.0	4.8	0.0	4.0	6.2	7.0*	5.0
Positive cocci	....	3.1	1.4	2.8	1.0	5.2	19.0	14.0	9.0	16.8	14.2	17.2	7.2	10.4	9.3
Oval free spores	....	4.2	3.2	3.8	7.0	1.2	0.6	1.0	2.8	1.6	1.2	1.8	2.2	0.6	2.4
Spherical free spores	....	1.8	10.0	1.8	1.4	1.2	0.4	0.4	0.6	0.0	0.2	0.6	0.0	0.4	1.4
Total Gram negative	....	88.0	83.8	88.8	87.4	84.8	75.2	78.4	79.6	76.8	78.4	76.4	84.4	81.6	81.9
Total Gram positive	....	6.0	3.0	5.6	4.2	12.8	23.8	20.2	17.0	21.6	20.2	21.2	13.4	17.4	14.3
Total free spores	....	6.0	13.2	5.6	8.4	2.4	1.0	1.4	3.4	1.6	1.4	2.4	2.2	1.0	3.8
Total negative rods	....	77.6	72.6	86.2	84.2	65.2	65.6	70.8	73.4	70.0	73.2	75.0	78.4	77.2	74.6
Total micrococci	....	13.3	11.2	5.4	3.4	24.2	28.4	21.2	15.0	22.8	19.4	18.6	13.2	14.8	16.2
Quality of the stain	....	....	....	....	....	....	....	....	....	....	Good	Fair	Fair	Good	

SUBJECT J.

Laboratory No.	B100	B111	B124	B136	B148	B159	B171	B183	B195	B207	B219	B231	B243	B255	Average
Date	Dec. 23	Jan. 9	Jan. 25	Feb. 10	Feb. 27	Mar. 13	Mar. 30	Apr. 14	Apr. 29	May 15	June 1	June 17	July 4	July 20	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type	51.8	68.2	Count not made	45.8	52.4	40.8	47.0	31.8	44.0	47.8	59.0	40.5	32.6	33.8	45.8
Other negative rods	21.2	3.2		21.2	38.6	40.0	31.8	34.6	31.4	27.4	22.2	30.0	35.6	21.2	27.6
Negative cocci	11.0	6.8		12.8	6.0	4.4	4.4	3.0	4.4	9.2	7.2	5.2	1.2	10.8	6.6
Negative spirochetes	2.4	0.8		10.6	0.4	1.0	0.4	3.4	0.6	0.2	0.4	0.5	4.2	11.2	2.8
Gram positive rods	5.6	8.6		3.8	1.4	1.6	2.8	4.2	6.2	4.6	2.6	4.8	7.2	6.2	4.6
Positive cocci	6.2	8.6		4.2	0.0	7.6	8.6	18.2	11.0	8.6	5.8	18.2	18.2	15.6	10.0
Oval free spores	1.2	0.8		1.2	1.2	3.4	4.8	2.4	1.6	0.6	2.0	0.4	0.4	1.2	1.7
Spherical free spores	0.6	3.0		0.4	0.0	1.2	0.2	2.4	0.8	1.6	0.8	0.4	0.6	0.0	0.9
Total Gram negative	86.4	79.0		90.4	97.4	86.2	83.6	72.8	80.4	84.6	88.8	76.2	73.6	77.0	82.8
Total Gram positive	11.8	17.2		8.0	1.4	9.2	11.4	22.4	17.2	13.2	8.4	23.0	25.4	21.8	14.6
Total free spores	1.8	3.8		1.6	1.2	4.6	5.0	4.8	2.4	2.2	2.8	0.8	1.0	1.2	2.6
Total negative rods	73.0	71.4		67.0	91.0	80.8	78.8	66.4	75.4	75.2	81.2	70.5	68.2	55.0	73.4
Total micrococci	17.2	15.4		17.0	6.0	12.0	13.0	21.2	15.4	17.8	13.0	23.4	19.4	26.4	16.7
Quality of the stain	....	....		....	....	Poor	Poor	Poor	Fair	Fair	Poor	Fair	Good	Good	

SUBJECT K.

Laboratory No.	....	B117	B129	B141	B151	B163	B175	B187	B199	B211	B223	B235	B247	B263	Average
Date	....	Jan. 16	Jan. 31	Feb. 19	Mar. 4	Mar. 19	Apr. 3	Apr. 20	May 4	May 20	June 5	June 23	July 9	July 29	
		Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type	....	51.0	37.4	40.2	40.2	44.7	51.6	58.0	50.2	Count not made	44.0	42.4	55.2	49.8	47.1
Other negative rods	....	20.0	19.8	19.2	20.6	19.3	18.4	17.4	24.4		26.6	32.4	22.8	25.4	22.2
Negative cocci	....	16.4	17.2	19.2	15.6	17.0	12.4	5.4	6.4		4.4	3.6	3.4	5.6	10.5
Negative spirochetes	....	0.2	0.2	1.4	1.2	1.2	0.8	0.4	0.2		0.0	0.0	0.4	9.4	1.3
Gram positive rods	....	4.8	11.2	3.0	8.6	5.1	5 6	7.2	8.8		11.8	7.4	6.0	4.8	7.0
Positive cocci	....	7.0	12.2	9.2	12.8	12.0	10.4	10.2	8.2		10.8	12.8	10.8	2.8	9.9
Oval free spores	....	0.4	0.2	0.6	0.2	0.0	0.4	0.8	0.4		2.2	1.0	0.6	1.6	0.7
Spherical free spores	....	0.2	1.8	7.2	0.8	0.7	0.4	0.6	1.4		0.2	0.4	0.8	0.6	1.3
Total Gram negative	....	87.6	74.6	80.0	77.6	82.2	83.2	81.2	81.2		75.0	78.4	81.8	90.2	81.1
Total Gram positive	....	11.8	23.4	12.2	21.4	17.1	16.0	17.4	17.0		22.6	20.2	16.8	7.6	16.9
Total free spores	....	0.6	2.0	7.8	1.0	0.7	0.8	1.4	1.8		2.4	1.4	1.4	2.2	2.0
Total negative rods	....	71.0	57.2	59.4	60.8	64.0	70.0	75.4	74.6		70.6	74.8	78.0	75.2	69.3
Total micrococci	....	23.4	29.4	28.4	28.4	29.0	22.8	15.6	14.6		15.2	16.4	14.2	8.4	20.5
Quality of the stain	....	....	....	....	Fair	Fair	....	Good	Good		Fair	Good	Good	Good	

SUBJECT L.

Laboratory No.	B108	B120	B132	B144	B155	B167	B179	B191	B203	B215	B227	B239	B251	B259	Average
Date	Jan. 2	Jan. 20	Feb. 3	Feb. 21	Mar. 9	Mar. 25	Apr. 8	Apr. 24	May 11	May 25	June 12	June 29	July 15	July 25	
	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent	Per cent
Bacillus coli type	46.2	81.6	50.2	46.8	40.6	53.4	52.8	58.0	54.8	58.6	48.6	57.6	46.2	45.0	52.9
Other negative rods	20.2	7.6	11.0	18.2	19.2	15.8	15.2	21.6	20.4	14.4	27.2	22.0	26.0	33.8	19.5
Negative cocci	20.2	3.6	20.6	11.8	11.6	18.6	13.6	8.0	3.4	3.8	0.8	3.6	7.0	4.2	9.3
Negative spirochetes	1.0	0.0	1.6	1.8	1.4	0.2	0.4	0.0	0.2	0.6	0.2	0.0	0.2	0.0	0.5
Gram positive rods	8.8	3.2	6.8	9.8	9.8	6.4	8.0	6.2	8.4	6.6	8.2	6.2	4.8	6.8	7.1
Positive cocci	2.6	2.2	8.6	9.0	16.0	3.2	9.4	5.0	11.0	14.2	13.6	9.8	14.6	9.0	9.2
Oval free spores	0.4	1.0	0.0	0.6	0.2	1.0	0.6	0.6	0.6	1.0	0.6	0.6	0.6	1.0	0.6
Spherical free spores	0.6	0.8	1.2	2.0	1.2	1.4	0.0	0.6	1.2	0.8	0.8	0.2	0.6	0.2	0.9
Total Gram negative	87.6	92.8	83.4	78.6	72.8	88.0	82.0	87.6	78.8	77.4	76.8	83.2	79.4	83.0	82.2
Total Gram positive	11.4	5.4	15.4	18.8	25.8	9.6	17.4	11.2	19.4	20.8	21.8	16.0	19.4	15.8	16.3
Total free spores	1.0	1.8	1.2	2.6	1.4	2.4	0.6	1.2	1.8	1.8	1.4	0.8	1.2	1.2	1.5
Total negative rods	66.4	89.2	61.2	65.0	59.8	69.2	68.0	79.6	75.2	73.0	75.8	79.6	72.2	78.8	72.4
Total micrococci	22.8	5.8	29.2	20.8	27.6	21.8	23.0	13.0	14.4	18.0	14.4	13.4	21.6	13.2	18.5
Quality of the stain	....	....	Good	Fair	Fair	Fair	Good	Poor	Fair	Fair	Good	Exc.	Exc.	Good	

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* One of these bacilli (0.2 per cent. of the bacteria counted) contained a spore.

were very rarely found; a single one is recorded in one count only, Subject C, May 23, 1908. As this is a unique observation, its significance must be regarded as doubtful. Very slender, undulating spirals we have classed as spirochetes. These measured from 3.5 to 6 μ in length; the thickness of the filament was about 0.25 μ, and the gross width of the coil from 0.5 to 0.9 μ. They were seen with great difficulty in unstained preparations but in a few instances where they happened to be especially numerous, they were seen and studied in the hanging drop. In the Gram stained preparations, and in the films stained with gentian violet for enumeration by the Eberle-Klein method, they were frequently observed. In some diarrheal stools these slender spirals were especially numerous. In these instances they appeared to be of the same kind as those ordinarily found in smaller numbers, but their specific identity with these is of course uncertain. The greatest number observed was 11.2 per cent of the fecal bacteria in Subject J on July 20, 1908. In many examinations none at all were found. The Gram positive bacilli varied from 13.0 per cent in Subject H, January 22, to 1.0 per cent in Subject G, July 29. Gram positive bacilli containing spores were seen from time time in five of the 12 subjects. The greatest number in any one examination occurred in Subject E, on March 2, 0.8 per cent of the bacteria counted. The Gram positive cocci varied from 31.6 per cent in Subject C, July 27, to 0.4 per cent in Subject A, February 6. Free spores were seen in nearly every examination. Oval free spores varied in quantity from 8.6 per cent in Subject A, May 28, to 0.0 in several instances. Spherical free spores were most numerous in Subject I, January 12, 10 per cent of the bacteria counted. In a number of examinations these also were absent.

The Gram stained films were always predominantly Gram negative. The total Gram negative bacteria, not including spores, varied between 97.4 per cent in Subject J, February 27, and 63.4 per cent in Subject G, March 4. The total Gram positive bacteria, including those containing spores, varied from 34.0 per cent in Subject C, July 27, to 1.4 per cent in Subject J, February 27. The total free spores was highest in Subject A, February 24, 13.6 per cent, and fell to 0.0 in Subject D, upon several occasions. This did not occur in any other subject however.

Gram negative bacilli composed the majority of the fecal bacteria in every case, varying from 91.0 per cent in Subject J, February 27, to 54.8 per cent in Subject B, February 13. The total micrococci, disregarding the staining reaction, varied from 39.4 per cent in Subject H, January 6, to 5.0 per cent in Subject C, March 23.

These data are summarized in Table 7, where the maxima, minima, and the means for each kind of bacteria for each subject, and the maximum, minimum, and mean of all the examinations are given.

SUMMARY. (PART I. DIRECT EXAMINATIONS.)

1. A homogeneous suspension of the bacteria of adult human feces may be readily prepared by making it sufficiently dilute, 1:100.

2. In such a suspension the bacteria can be counted microscopically by an experienced observer, with a fair degree of accuracy.

3. The gravimetric method of Strasburger for determining the quantity of fecal bacteria is more time-consuming, but the method is capable of a higher degree of accuracy than the enumeration procedures. The separation of the bacteria should be done by fractional sedimentation in a high-speed centrifuge, and requires careful work at every step.

4. In the case of normal adult men, eating an ordinary mixed diet, the average number of fecal bacteria excreted daily is about 33×10^{12}; the average daily bacterial dry substance about 5.34 gm., and the daily bacterial nitrogen 0.585 gm., making up 46.3 per cent of the total fecal nitrogen. There is considerable individual variation in the average quantity of fecal bacteria even in persons taking the same diet.

5. The results of enumeration do not correspond accurately with the results of gravimetric determination of the bacteria under different conditions. The fecal bacteria as individuals contain an amount of dry substance subject to considerable variation.

6. The bacteria of the adult human feces are Gram negative for the most part, about 70 per cent of all the bacteria being Gram negative bacilli. Gram positive rods are constantly present.

7. It is possible to recognize microscopically a number of morphologically different bacteria, present in such numbers that they must have resulted from multiplication in the intestine. A quantity of bacteria sufficient to yield 0.5 gm. or even 0.1 gm. of dry bacterial sub-

TABLE 7

Summary of Differential Counts of Fecal Bacteria.

		Subject A	Subject B	Subject C	Subject D	Subject E	Subject F	Subject G	Subject H	Subject I	Subject J	Subject K	Subject L	All Subjects
Bacillus coli type	Maximum	62.6	44.4	67.0	67.6	63.2	53.4	63.2	58.4	69.8	68.2	58.0	81.6	81.6
	Mean	**4 .4**	**39.1**	**48.2**	**45.8**	**43.0**	**44.0**	**45.1**	**47.0**	**40.2**	**45.8**	**47.1**	**52.9**	**45.9**
	Minimum	32.8	31.6	33.4	28.4	19.0	37.8	30.0	32.8	34.0	31.8	37.4	40.6	19.0
Other negative rods	Maximum	37.8	36.6	35.2	40.0	46.2	32.4	32.3	35.4	44.4	40.0	32.4	27.2	46.2
	Mean	**27.2**	**28.7**	**25.6**	**24.0**	**31.3**	**22.9**	**25.0**	**24.2**	**25.4**	**27.6**	**22.2**	**19.5**	**25.3**
	Minimum	12.3	17.6	8.2	2.0	8.2	14.6	16.0	0.4	3.6	3.2	17.4	7.6	0.4
Negative cocci	Maximum	12.8	26.2	10.8	22.4	13.2	21.4	11.8	36.8	19.0	12.8	19.2	20.6	36.8
	Mean	**5.6**	**10.8**	**5.2**	**9.9**	**7.6**	**10.7**	**7.5**	**11.2**	**6.9**	**6.6**	**10.5**	**9.3**	**8.4**
	Minimum	1.4	2.2	1.2	3.0	3.8	3.8	3.2	3.6	1.4	1.2	3.4	0.8	0.8
Negative spirilla	Maximum	0.0	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2
	Mean	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**
	Minimum	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Negative spirochetes	Maximum	3.2	3.8	5.2	7.6	2.0	1.0	3.4	1.8	1.4	11.2	9.4	1.8	11.2
	Mean	**0.4**	**0.8**	**1.8**	**2.4**	**0.5**	**0.3**	**1.1**	**0.6**	**0.4**	**2.8**	**1.3**	**0.5**	**1.1**
	Minimum	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.2	0.0	0.0	0.0
Gram positive rods	Maximum	11.9	7.2	11.4	11.0	7.8	12.0	11.2	13.0	8.0	8.6	11.8	9.8	13.0
	Mean	**6.2**	**4.0**	**4.4**	**7.4**	**3.8**	**8.4**	**5.2**	**4.8**	**5.0**	**4.6**	**7.0**	**7.1**	**5.7**
	Minimum	3.8	2.4	2.2	3.6	1.6	2.2	1.0	1.8	1.6	1.4	3.0	3.2	1.0
Positive rods with spores	Maximum	0.2	0.2	0.0	0.0	0.8	0.0	0.6	0.0	0.2	0.0	0.0	0.0	0.8
	Mean	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.2**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**
	Minimum	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Positive cocci	Maximum	19.4	26.8	31.6	18.0	24.2	19.2	26.0	18.4	17.2	18.2	12.8	16.0	31.6
	Mean	**12.4**	**15.9**	**13.1**	**9.6**	**11.2**	**12.0**	**12.5**	**10.5**	**9.3**	**10.0**	**9.9**	**9.2**	**11.3**
	Minimum	0.4	3.2	2.6	3.4	1.2	7.8	4.0	2.6	1.0	4.2	2.8	2.2	0.4
Oval free spores	Maximum	8.6	0.8	3.4	0.8	2.2	1.8	4.4	3.0	7.0	4.8	2.2	1.0	8.6
	Mean	**4.1**	**0.4**	**1.3**	**0.4**	**1.3**	**0.9**	**2.4**	**1.4**	**2.4**	**1.7**	**0.7**	**0.6**	**1.4**
	Minimum	0.0	0.0	0.3	0.0	0.4	0.2	1.0	0.0	0.6	0.4	0.0	0.2	0.0
Spherical free spores	Maximum	8.6	1.2	2.2	2.0	2.8	1.4	4.4	0.8	10.0	3.0	7.2	2.0	10.0
	Mean	**1.7**	**0.3**	**0.4**	**0.5**	**0.7**	**0.8**	**1.2**	**0.3**	**1.4**	**0.9**	**1.3**	**0.9**	**0.9**
	Minimum	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0
Total Gram negative	Maximum	89.6	91.4	93.4	90.4	93.8	87.4	93.0	95.6	88.8	97.4	90.2	92.8	97.4
	Mean	**75.6**	**70.3**	**80.8**	**82.1**	**83.0**	**77.9**	**78.7**	**83.0**	**81.9**	**82.8**	**81.1**	**82.2**	**80.7**
	Minimum	69.4	68.4	65.0	75.6	70.0	72.0	63.4	76.2	75.2	73.6	74.6	72.8	63.4
Total Gram positive	Maximum	26.9	30.8	34.0	26.4	28.4	29.0	33.2	28.8	23.8	25.4	23.4	25.8	34.0
	Mean	**18.6**	**20.0**	**17.5**	**17.0**	**15.0**	**20.4**	**17.7**	**15.3**	**14.3**	**14.6**	**16.9**	**16.3**	**17.0**
	Minimum	4.2	7.4	5.2	7.8	2.8	10.2	5.0	2.6	3.0	1.4	7.6	5.4	1.4
Total free spores	Maximum	13.6	1.6	5.6	2.2	5.0	2.4	7.0	3.2	13.2	5.0	7.8	2.6	13.6
	Mean	**5.8***	**0.7**	**1.7**	**0.9**	**2.0**	**1.7**	**3.6**	**1.7**	**3.8**	**2.6**	**2.0**	**1.5**	**2.3**
	Minimum	0.2	0.2	0.5	0.0	0.8	0.4	1.7	0.2	1.0	0.8	0.6	0.6	0.0
Total negative rods	Maximum	80.2	78.8	87.0	77.4	88.0	78.4	85.2	84.8	86.2	91.0	78.0	89.2	91.0
	Mean	**60.6**	**67.8**	**73.8**	**69.8**	**74.9**	**66.9**	**70.1**	**70.1**	**74.6**	**73.4**	**69.3**	**72.4**	**71.1**
	Minimum	60.1	54.8	62.4	63.2	64.0	55.2	59.2	56.8	65.2	55.0	57.2	59.8	54.8
Total micrococci	Maximum	31.6	36.0	32.8	26.8	30.2	32.0	31.2	39.4	28.4	26.4	29.4	29.2	39.4
	Mean	**18.0**	**25.9**	**18.3**	**10.5**	**18.8**	**22.7**	**19.0**	**21.7**	**16.2**	**16.7**	**20.5**	**18.5**	**19.7**
	Minimum	8.2	14.2	5.0	12.2	8.2	14.8	10.6	10.4	3.4	6.0	8.4	5.8	3.4

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* Note in this connection that Subject A excreted the driest feces of all the twelve subjects.

stance can hardly have been taken in as such with the food. Therefore there are several species of bacteria whose normal habitat is the human intestine.

8. Free spores are almost constantly present in considerable numbers in the feces. They are more numerous in dry stools as a rule. Diarrheal stools, however, sometimes show a very large number of free spores.

9. Thin, flexible spirals are quite frequently present in normal stools, and under some conditions are very numerous.

10. The direct quantitative determinations of the fecal bacteria furnish evidence of the extent and nature of the bacterial growth in the intestine. This seems to be a delicate index of intestinal conditions.

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